



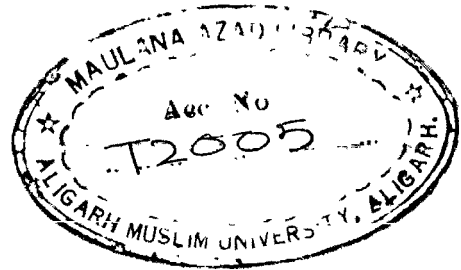
STUDIES ON HADRONCHUS SHAKILI JAIRAJPURI, 1969  
( NEMATODA : MONONCHIDA )

**ABSTRACT**

THESIS SUBMITTED TO THE  
ALIGARH MUSLIM UNIVERSITY, ALIGARH  
FOR THE AWARD OF THE DEGREE  
OF  
Doctor of Philosophy  
IN  
ZOOLOGY

BY  
**NASEEM AHMAD**

DEPARTMENT OF ZOOLOGY  
ALIGARH MUSLIM UNIVERSITY,  
ALIGARH  
MARCH, 1980



## ABSTRACT

10 DEC 1980

The predatory nematode, Hadronchus shakili Jairajpuri, 1969 is a large sized mononch widely distributed in this country especially in the Northern and North-eastern regions. The present work on this nematode species includes study of its gross morphology and histological anatomy and the description of its various juvenile stages. The variability of important taxonomic characters in the adults and the juveniles has been statistically analysed. Comparative development of various organs in juveniles and adults has also been studied in detail followed by observations on the population dynamics of the species. Lastly, the effects of some toxic chemicals were determined.

The morphological studies revealed that the shape of body, lip region and buccal cavity show little variations but the total body length, number of denticles on the subventral walls, vulval papillae, number of ventromedian supplements, tail length etc. are more variable. The size of the buccal cavity is to some extent correlated with the sex and length of the body of the animal.

The anatomical observations were carried out with particular reference to musculature, digestive and reproductive organs. The cross sections at various levels of the body show

four lateral chords and four platymarian somatic muscle cells per quadrant. Well developed labial muscles are attached to <sup>the</sup> lips and stomatal muscles to the stoma. The vulval muscles are also of two types, the constrictor vulvae and the dilator vulvae. The oviduct-uterus junction is provided with prominent sphincter muscles. The copulatory muscles, accessory copulatory muscles, spicular muscles, and caudal copulatory muscles are also well developed. In females, the anal region has 2-3 anal muscle bands while in males there is a single band. The digestive organs include hexaradiate vestibulum, tri-radial buccal cavity, muscular oesophagus and tuberculate type of oesophago-intestinal junction. The lumen of oesophagus is tri-radiate and heavily sclerotized, the thickenings serving as points of attachment for the radial muscle fibers. The intestine has 6-8 cells in circumference. The females are amphidelphic and each sexual branch consists of a reflexed ovary, an oviduct with narrow distal and an enlarged sac-like proximal part and a long flexible uterus. The males are diorchic, each testis is followed by a vas deferens, and an ejaculatory duct joining the cloaca. The copulatory structures include paired spicules, a gubernaculum and lateral accessory pieces.

The important characters for separating the different juvenile stages are, total body length; length and width of

buccal cavity; number, orientation and position of teeth in the buccal cavity; length of oesophagus; size of the genital primordia etc. The first stage juveniles are always devoid of denticles on the vertical subventral walls while in the remaining stages these are present. The juveniles can easily be distinguished from their adults by the presence of a spare tooth at the base of the functional dorsal tooth.

The analysis of variability of some important morphological characters of the adults and juveniles revealed that almost all the characters are variable to some extent. The length and width of buccal cavity are, however, least variable. The position of nerve ring, anal body-width and length of spicules are also not much variable. The highly variable characters include the position of amphid apertures, length of female sexual branches, tail, rectum etc. The vulva position is positively correlated with the total body length while the lengths of genital branches are negatively correlated. Of all the allometric characters, the value of  $V$  is least variable and  $G_1$  and  $G_2$  exhibit maximum variability. The length of buccal cavity, oesophagus and tail are independent of body length. The characters in juveniles vary almost to the same extent as in the adults.

The buccal cavity is formed anew during the process of moulting. A total of four moults occur. In the late phase of

each moult there are always three dorsal teeth one of these is cast off along with the old buccal cavity, the second one becomes the functional tooth of the stage to follow, while the third one is the spare tooth of the future stage. The development of the buccal cavity is more pronounced in the adults than in the juveniles. The development of oesophagus and tail in relation to total body length is more in juveniles than the adults. The genital primordia in the first stage juveniles consists of two oval bodies connected by a cellular strand. Each oval body is with one germinal nucleus and two somatic nuclei. The cap cell nuclei are derived from the division of the somatic nuclei at the distal and proximal ends of the primordia and do not divide again. The germinal nuclei divide for the first time in the third moult while the somatic nuclei in the second moult. The somatic nuclei proliferate in the mixxle region of the primordia to form the gonoduct. The epidermal nuclei are derived from the somatic nuclei at the opposite ends of the primordia and form the epidermal walls of the testes and ovaries. The vagina is formed by the specialized ventral chord nuclei, whereas the development of spicules and gubernaculum takes place from the spicular primordia in the anal region, <sup>which is</sup> distinguishable in the third stage male juveniles. The sex differentiation occurs in the second stage juveniles when specialized ventral chord nuclei make their appearance in the female juveniles. The development

of the genital tract and ovary in various seasons also differed. The frequency of egg production by the anterior ovary was much higher than that of the posterior ovary (1:2).

Observations were made on the fluctuations in the populations of this nematode in a plot of about one acre at the Company Gardens, Bareilly, Uttar Pradesh from July 1977 to June 1978. For this purpose, soil samples at depths of 0-10, 10-20 and 20-30 cm were collected twice a month. The nematodes were extracted separately from these three types of samples and were counted and classified age-wise. The seasonal fluctuations in the population was found to be chiefly governed by the moisture contents of the soil, because the maximum level of the population was noted during the peak monsoon period (August) while the minimum during the dry period (April). The data on the vertical migration revealed that this species occurs abundantly in the surface layer from May to October (Except for a brief period during July when the entire field was water-logged). From October onwards they start to migrate downwards and concentrate at 10-20 cm depth until April. At the depth of 20-30 cm the population was always found to be low. On the basis of abundance of juveniles and the gravid females, the breeding period of this nematode appears to begin in March and extends up to October, but apparently they bred only once a year. An inverse relationship exists between the population of

this mononch and those of its prey, the soil-inhabiting nematodes including plant parasites. This suggests a clear antagonism between the prey and predator.

The observations on the effects of pH, mineral salts and fatty acids showed that these chemicals are toxic to this species. The high susceptibility to the pH gradients ranging 2.2 to 4.0 and 6.4 to 8.0 showed that these nematodes can not survive either in high acidic media or low alkaline media. The maximum life span at pH 5.8 to 6.0 shows that this is the optimum pH range. The mineral salts of concentrations ranging from 0.4M to 0.05M showed different effects at different concentrations. The rate of survival was lower at higher concentrations. Copper sulphate was most toxic, while potassium chloride was least. The fatty acids ranging from 1N to 0.001N also proved highly toxic, but the toxicity at lowest concentration of 0.001N was less. Formic acid proved highly toxic. The adults in general were more tolerant than the juveniles. The adult females were least susceptible while the first stage juveniles were most. The exposure to various chemicals also brought about changes in the posture and movement of the nematodes.



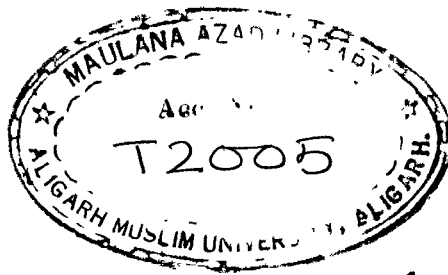
**STUDIES ON HADRONCHUS SHAKILI JAIRAJPURI, 1969**  
**( NEMATODA : MONONCHIDA )**

THESIS SUBMITTED TO THE  
ALIGARH MUSLIM UNIVERSITY, ALIGARH  
FOR THE AWARD OF THE DEGREE  
OF  
**Doctor of Philosophy**  
IN  
**ZOOLOGY**

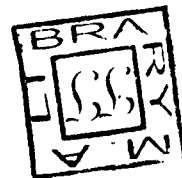
BY  
**NASEEM AHMAD**

DEPARTMENT OF ZOOLOGY  
ALIGARH MUSLIM UNIVERSITY,  
ALIGARH  
MARCH, 1980





20 DEC 1980



T2005

*St*  
CHECKED-2002


M. Shamim Jairajpuri  
M.Sc., Ph.D., D.Sc. F.Z.S.I.  
President; Nematological Society of India

DEPARTMENT OF ZOOLOGY  
ALIGARH MUSLIM UNIVERSITY  
ALIGARH - 202 001, INDIA

22.3.1980

SUPERVISOR

This is to certify that the entire research work presented in the thesis entitled "Studies on Hadronchus shakili Jairajpuri, 1969 (Nematoda : Mononchida)" by Mr. Naseem Ahmad is original and was carried out under my supervision. I have allowed Mr. Ahmad to submit it to the Aligarh Muslim University in fulfilment of the requirements for the degree of Doctor of Philosophy in Zoology.

  
(M. Shamim Jairajpuri)  
Reader-in-Charge  
Section of Nematology.

## ACKNOWLEDGEMENTS

The author is deeply indebted to Dr. M. Shamim Jairajpuri for supervising the work and for critically going through the manuscript.

Thanks are also due to Prof. S. Mashhood Alam, Head of the Department of Zoology, Aligarh Muslim University for providing necessary facilities. I am also deeply grateful to Dr. Qaiser Husain Baqri for his help and suggestions. The co-operation of colleagues of the Nematology Research Laboratory during the entire period of the study has been note-worthy and is acknowledged here with thanks.

The financial assistance from the Indian Council of Agricultural Research, New Delhi is also gratefully acknowledged.

# THESIS SECTION

## CONTENTS

	Page
INTRODUCTION      ...      ...      ...      ...	1
MATERIAL AND METHODS      ...      ...      ...	5
GROSS MORPHOLOGY      ...      ...      ...	8
HISTOLOGICAL ANATOMY      ...      ...      ...	16
JUVENILE STAGES      ...      ...      ...	32
VARIABILITY      ...      ...      ...	37
COMPARATIVE DEVELOPMENT      ...      ...      ...	54
POPULATION DYNAMICS      ...      ...      ...	69
EFFECTS OF CHEMICALS      ...      ...      ...	79
SUMMARY      ...      ...      ...	93
REFERENCES      ...      ...      ...	99
TABLES I - VIII      ...      ...      ...	117
FIGURES I - XXXVII      ...      ...      ...	127

## INTRODUCTION

The nematodes are one of the largest groups of animals. Those which live in soil are either free-living, saprophytic, phyto-parasitic or predatory. The economic importance of phyto-parasitic nematodes is now well established because of the fact that they feed upon plants and cause extensive damage. Recently some attention has also been paid to the predatory nematodes which may play a role in the biological control of plant-parasitic nematodes. Among the most important predators are those belonging to the order Mononchida Jairajpuri, 1969 which are commonly referred to as mononchs.

The observations on the feeding habits of some species of mononchs have revealed that either they engulf their prey, or cut them into pieces or suck the contents of their bodies. Steiner and Heinly (1922) who worked on the feeding habits of Clarkus papillatus found that a large number of root-knot nematodes could be killed by each of these predators in one day. During its life span of 12 weeks, a single mononch could kill 1322 nematodes. Cobb (1920) noticed that C. papillatus could bring down the population of sugar beet nematodes to a low level. Thorne (1927) also found C. papillatus to be useful in the control of sugar beet nematodes, Heterodera schachtii, provided a large population of the mononch was present in the infested fields.

Cassidy (1931) observed Iotonchus brachylaimus devouring large numbers of Heterodera eggs and larvae in cultures. Mulvey (1961) observed C. papillatus swallowing its prey while Anatonchus tridentatus devouring a nematode. Esser (1963) gave an account of feeding behaviour in mononchs. Nelmes (1972) discussed cultivation and feeding behaviour of Prionchulus punctatus. Cohn and Mordechai (1974) used Mylonchulus sigmaturus as predator on Tylenchulus semipenetrans. Recently, Jairajpuri and Azmi (1978) have worked on the predatory behaviour of Mylonchulus dentatus, a species which commonly occurs in India.

Several workers have reported and described a large number of species of mononchs from different parts of the world (Cobb, 1916 & '17; Clark, 1960-'63; Mulvey, 1961-'78; Mulvey and Jensen, 1967; Jairajpuri, 1969-'71 etc. etc.). Most of these studies are confined only to the taxonomy of Mononchida and very little work has been done on the anatomy, variability, biology, ecology etc. of these animals. Cobb (1916 & '17) provided a good account of the morphology, biology and taxonomy of mononchs. Coomans and Lima (1965), Coomans and Loof (1970) and S. Z. Baqri and Jairajpuri (1974) contributed much to our existing knowledge of morphology of this group. Very recently, Grootaert and Wyss (1979) have studied the fine structures of the anterior feeding apparatus of Mononchus aquaticus and have also discussed the feeding mechanism and functions of various types of muscles which

control the feeding apparatus. Keeping in view the possible economic importance of mononchs and the pioneering work done on this group from our Department of Zoology of the Aligarh Muslim University, the present work was undertaken. It provides in detail the morphology, anatomy, variability, development and the ecology of a species of predatory nematode, Hadronchus shakili Jairajpuri, 1969. This is large sized species and is an efficient predator on other type of nematodes and is known to occur in different regions of this country.

The genus Hadronchus was erected by Mulvey and Jensen (1967) who described two species, viz., the type H. bisexualis and H. monohystera from Nigeria. Jairajpuri (1969) described two more species, viz., H. andamanicus and H. shakili from India. Thong (1971) added H. yueni from Singapore. Recently, Mulvey (1978) proposed the genus Parahadronchus for three species of Hadronchus i.e., H. andamanicus Jairajpuri, 1969, H. shakili Jairajpuri, 1969 and H. yueni Thong, 1971. He differentiated this genus from Hadronchus in the position of dorsal tooth, number and location of denticles, presence of caudal glands and in the shape of the tail. In the present work instead of using the name Parahadronchus shakili (Jairajpuri, 1969) Mulvey, 1978, the old name Hadronchus shakili has been used.

The work included in this thesis has been dealt under nine chapters. The study is based mostly on the specimens from

a natural population collected at Company Gardens, Bareilly. To begin with, a description of the species, Hadronchus shakili has been provided followed by detailed observations on the anatomy, in particular of musculature, digestive and reproductive system etc. The juveniles obtained from natural population were classified into four stages and all these stages have been adequately described and figured. A detailed discussion on the variability of different structures more particularly those of taxonomic importance have been provided. This is followed by comparative development of various organs of this nematode species. Observations on the population dynamics reveal that the level of population of this nematode fluctuates during the different seasons of the year and is dependent upon moisture content, temperature etc. The species breeds only once a year but the breeding period extends from March to October. The predatory ability of H. shakili was quite evident from the fact that slight rise in its population brought about distinct decline in the population level of its prey (other nematodes). Effect of some chemicals and pH on this nematode was also observed.



## MATERIAL AND METHODS

### Soil Sampling:

The soil samples were collected at a depth of 6 to 10 inches. These were kept in polythene bags and brought to the laboratory for isolation of nematodes.

### Processing of samples:

About 500 gm of soil was taken into a bucket which was filled with water to about 1/3rd of its volume. The soil was thoroughly mixed with water and left undisturbed for a few seconds so as to allow the heavy soil particles to settle down at the bottom of the bucket. It was then passed through a coarse sieve in order to get rid of undesirable substances like roots, leaves, debris and other organic matters which accumulated on the top of the sieve while the filtrate was collected in another bucket. This process was repeated thrice. Afterwards the muddy suspension in the bucket was stirred gently and then passed through a sieve of mesh No. 300, pore size 53  $\mu$ m. The nematodes along with some soil particles remain on the sieve. The residue thus collected was transferred to a beaker. The same process was repeated 3 to 4 times for maximum recovery.

### Isolation:

The residue from the beaker was poured on tissue paper placed on a small coarse nylon sieve which was placed on a conical

## MATERIAL AND METHODS

### Soil Sampling:

The soil samples were collected at a depth of 6 to 10 inches. These were kept in polythene bags and brought to the laboratory for isolation of nematodes.

### Processing of samples:

About 500 gm of soil was taken into a bucket which was filled with water to about 1/3rd of its volume. The soil was thoroughly mixed with water and left undisturbed for a few seconds so as to allow the heavy soil particles to settle down at the bottom of the bucket. It was then passed through a coarse sieve in order to get rid of undesirable substances like roots, leaves, debris and other organic matters which accumulated on the top of the sieve while the filtrate was collected in another bucket. This process was repeated thrice. Afterwards the muddy suspension in the bucket was stirred gently and then passed through a sieve of mesh No. 300, pore size 53  $\mu$ m. The nematodes along with some soil particles remain on the sieve. The residue thus collected was transferred to a beaker. The same process was repeated 3 to 4 times for maximum recovery.

### Isolation:

The residue from the beaker was poured on tissue paper placed on a small coarse nylon sieve which was placed on a conical

funnel filled with water barely touching the bottom of the sieve. The nematodes migrate through the filters into the clean water of funnel and settle down at the bottom of the funnel. After about 24 hours the nematodes along with a small amount of water were collected in a test tube or cavity block. The presence of nematodes was also determined in the residue on the filter.

#### Killing and fixing:

The nematodes were killed and fixed in 4% formaldehyde. For this purpose the suspension was allowed to settle down for 2-4 hours. The excess water was discarded and the nematodes were left in a very small amount of water. To this an equal amount of hot 4% formaldehyde was poured which killed and fixed them instantaneously. The nematodes could be stored in this medium indefinitely.

#### Mounting and sealing:

The nematodes were transferred to a mixture of glycerine and 30% alcohol (glycerine 5 parts: alcohol 95 parts), kept in a small cavity block. The block was put in a desiccator at room temperature for 2 to 3 weeks. The alcohol evaporated leaving the nematodes in pure glycerine. The dehydrated nematodes were mounted in anhydrous glycerine on glass slides. Pieces of glass wool of adequate thickness were always placed between the slide and coverslip to check the pressure on the specimens. The mounts were sealed either with 'cutex' neutral colour nail polish

or 'Glyceel'.

Cross sections of Nematodes:

The transverse sections of nematode body at various levels were cut with the help of a sharp razor blade in glycerine-jelly and were mounted in the same medium.

Measurements and Drawings:

The nematodes were examined and measured with the help of an ocular micrometer. For denoting measurements the de Man's formula (1884) was used. The drawings were made with the help of Camera Lucida. In the text  $\mu\text{m}$  stand for  $\mu\text{m}$ .

## GROSS MORPHOLOGY

The description of Hadronchus shakili was originally provided by Jairajpuri (1969) based on 14 specimens from Nainital, Bareilly and Gola Gokarnath, Uttar Pradesh. The following description is based on the study of a very large number of specimens collected mainly from Company Gardens, Bareilly (Uttar Pradesh) during different periods of the year. Two other populations, one from Sikkim and the other from Malaysia were also studied but there were few specimens in these populations. With this wealth of material at hand it has been possible to study in detail the gross morphology of this species and also to determine the extent of variability of various characters.

Dimensions: Table I.

Description:

Female: Worms are whitish in colour and have long bodies which assume a ventrally arcuate posture upon fixation and tapering slightly anteriorly, but markedly posteriorly. Cuticle is 3-6  $\mu$ m thick at various places on the body. Lateral chords  $1/6$ th -  $1/3$ rd of the corresponding body-width near the middle in Bareilly population,  $1/6$ th -  $1/5$ th in the Sikkim and Malaysia populations.

The lip region is wider than adjoining body, distinctly offset. The range for lip-width and lip height differs in the

Bareilly and Sikkim populations, 45-60 x 15-25 um and 34-39 x 11-14 um respectively; while in Malaysia population it is 44-46 x 15-17 um. En face view (Fig. X, A) shows the vestibulum bounded by six lips which are of similar shape and size. There are two dorsolateral, two ventrolateral and two lateral lips. Each lip is provided with an outer and an inner papilla. Thus the two circlets have six papillae each. Near the middle of lips on either side of internal papillae are present prominent 'dot' like structures. Amphids cup-shaped, 5-8 um wide, 14-25 um from anterior end. The amphidial chamber is connected to an amphidial duct which leads to the sensillar pouch containing sensory neurons.

Stoma consists of a hexaradiate vestibulum and a barrel-shaped tri-radiate buccal cavity. Walls of buccal cavity are formed by two sets of heavily cuticularized plates, the ventral and the oblique sets. The dorsal wall of the vertical set bears a large tooth in the posterior half of buccal cavity. The subventral walls bear teeth and the oblique walls have two foramina each. The range of the length and width of buccal cavity in Bareilly, Sikkim and Malaysia population is 50-65 x 32-40 um (60 x 36 um), 44-46 x 25-28 um (45 x 26 um) and 54-56 x 31-33 um (55 x 32 um) respectively. Distinct variations were noticed in the position of dorsal tooth which is situated at 37-47% (43%) in Bareilly and Sikkim populations whereas at 54-55% in the Malaysia population from base of buccal cavity.

The Malaysia population further differs in shape of dorsal tooth which is more thicker and stouter. The number of denticles on the subventral walls is variable in all the three populations, 2-7 in Bareilly and Malaysia populations; 2-5 in Sikkim population.

Oesophagus is roughly cylindrical and strongly muscular, with tri-radiate lumen, slightly expanded at its anterior end, narrowing a little at nerve ring and then widening towards base. The oesophago-intestinal junction is tuberculate with non-sclerotized tri-radiate lumen. Though the oesophagus varies in all the three populations, 476-716  $\mu\text{m}$  (662  $\mu\text{m}$ ) in Bareilly, 472-483  $\mu\text{m}$  (477  $\mu\text{m}$ ) in Sikkim and 446-626 <sup>$\mu\text{m}$</sup>  (636  $\mu\text{m}$ ) in Malaysia but the value of 'b' is almost same. The nerve ring is located at 145-198  $\mu\text{m}$  or 24-32% (28%) in Bareilly population, 164-166  $\mu\text{m}$  or 26% in Malaysia population and 108-144  $\mu\text{m}$  or 28-32%(30%) of oesophageal length from the anterior end in Sikkim population. The location of orifices of oesophageal glands from anterior end is almost same in Bareilly and Malaysia populations being 41-45% (43%), 59-64% (61%) and 85-90%(89%) for  $\text{DO}$ ,  $\text{S}_1\text{O}$  and  $\text{S}_2\text{O}$  respectively. In Sikkim population these positions are as follows:  $\text{DO}$  = 49-53% (50%);  $\text{S}_1\text{O}$  = 66-69%(68%);  $\text{S}_2\text{O}$  = 91-92% (91%). The nuclei of these glands could not be seen.

Vulva is transverse, slit-like, post-equatorial with cuticularized lips. Vulval papillae are present on either side

of vulva. Of the 140 females observed from Bareilly population, they varied from being completely absent on either side of vulva to a maximum of four prevulval and three postvulval. Between these two extremes many variations were found (Fig. III, Table II). In Sikkim population the females were noted with one pre- and one postvulval papilla while in Malaysia population their number varied from 1-2 prevulval and one postvulval. Not only that their number is variable, but also their position from vulva and from one another is quite variable. Moreover, their state of development varies from being fully developed to a very poorly developed. Vagina short, thick-walled. Gonads amphidelphic, ovaries reflexed. Each sexual branch consists of an ovary, an oviduct with a narrow distal and an enlarged sac-like proximal part, well developed sphincter and a large flexible uterus. The sexual branches show much variability in their lengths (Fig. IV and V). The length of the ovary also shows wide range of variations, from poorly developed having only few oocytes (Fig. V, E) to more than half of the length of normal genital branch (Fig. V, B). The shape of the uterine egg is oval to elongate-oval, its size usually varies from 124-150 x 51-62  $\mu\text{m}$  (140 x 48  $\mu\text{m}$ ). However, in some abnormal cases the size may be as small as 102 x 48  $\mu\text{m}$  and as large as 198 x 62  $\mu\text{m}$ . Apart from normal gonads, some abnormalities were noticed which are described below: i) In one female along with normal gonads an additional reproductive system very poorly developed having



both the branches is present dorsal to anterior reproductive branch. A cuticular protuberance is present indicating a primitive vulva formation. In addition two 'dot'-like structures are also present which may be the poorly developed cuticular pieces (Fig. VI, A). ii) In another female, an additional anterior sexual branch is present. This branch consists of all the normal reproductive parts but is smaller in length and has a shorter ovary (Fig. VI, B). iii) A female having outstretched anterior ovary (Fig. VI, C). iv) In another female both the sexual branches are located posterior to vulva which resulted in a pseudo-di-opisthodelphic condition (Fig. VI, D).

The tail length measures 262-430  $\mu\text{m}$  (372  $\mu\text{m}$ ) or 6-10(8) anal body-widths long in Bareilly population (Fig. VII, A & B), 387-445  $\mu\text{m}$  (408  $\mu\text{m}$ ) or about 10-12 (11) anal body-widths long in Sikkim population (Fig. VII, C & D) and 578-600  $\mu\text{m}$  (589) or about 13-14 anal body-widths long in Malaysia population (Fig. VII, E & F).

Male: The males have been recorded only from Bareilly and Sikkim populations. They resemble females in all the morphological characters except that they have strongly curved body in posterior half.

Buccal cavity:

Buccal cavity measures 49-60 x 29-35 um (55 x 31 um) in Bareilly population, 40-42 x 22-26 um (41 x 23 um) in Sikkim population. The dorsal tooth is situated at 38-46% in Bareilly population, 38-44% in Sikkim population from base of buccal cavity. The length of spicules along the median line differs in both the populations, 71-79 um (74 um) and 91-106 um (96 um) in Sikkim and Bareilly populations respectively.

The gubernaculum is 18-24 um (21 um) long in Bareilly population whereas in Sikkim population it is 15-20 um (17 um) long; lateral accessory pieces are 12-18 um (13 um) in Bareilly population and 11-14 um (12 um) long in Sikkim population. The number of ventromedian supplements in Bareilly population varies from 11 to 16. Upon analysis it was noted that 11 ventromedian supplements were present in 9%, 12 in 14%, 13 in 57%, 14 in 6%, 15 and 16 only in 1% specimens. All the males from the Sikkim population had 11 ventromedian supplements.

The tail length is also highly variable, measures 245-335 um (297 um) or 5-7 (6) anal body-widths long in Bareilly population (Fig. VIII, D & E) and 248-379 um (341 um) or 6-8 (7.5) anal body-widths long in Sikkim population (Fig. VIII, F & G). In one male specimen the tail is 165 um long with slightly broader tip (Fig. VIII, A), in another specimen the tail measures 183 um with more developed caudal glands and rounded terminus (Fig. VIII, B).

In yet another male the tail is 200 um in length (Fig. VIII, C).

#### DISCUSSION

The above observations show that morphological variations are exhibited not only in the populations from different geographical locations but also in a single population. The shape of body, lip region and buccal cavity are not greatly variable, but the total body length, width and height of lips, length and width of buccal cavity within and among different populations vary to some extent. The shape as well as the position of dorsal tooth does not vary much within a population but it is highly variable among different populations. Variations were also noticed in the orifices of the oesophageal glands. The female reproductive system shows a high degree of variability and its careful study seems useful because it provides important taxonomic characters. The number of vulval papillae vary from 0-4 and their position also shows wide range of variations. The tail also shows much variability. The important differences between the three populations of this species are given in the following Table (III).

In spite of the differences, as are evident from the Table, the three populations cannot be separated at species level because of the intra-morphological agreements. These differences should be considered intra-specific variations. The present study,

therefore, reveals that H. shakili is an extremely variable species.

TABLE - III

Characters	POPULATIONS		
	Bareilly	Sikkim	Malayasia
Buccal cavity	49-65 x 29-40 um	40-46 x 22-28 um	54-56 x 31-33 um
Dorsal tooth (from base of buccal cavity)	Less developed 37-47%	Less developed 38-47%	Well developed 54-55%
DO	41-45%	49-53%	42-43%
S <sub>1</sub> O	59-64%	66-69%	61-62%
S <sub>2</sub> O	85-90%	91-92%	89%
'V'	60-66%	56-57%	56-57%
'Va'	65-68%	70-72%	70-72%
Length of spicules	91-106 um	71-79 um	-
Gubernaculum	18-24 um	15-20 um	-
Lateral accessory pieces	12-18 um	11-14 um	-
Tail/anal body width	OO ++ ↑↑ OO	6-10 um 10-12 6-8	13-14 -

Detailed observations on variability in adults and juvenile stages and its statistical analysis is given in the chapter entitled 'Variability' from page 37-53.

## HISTOLOGICAL ANATOMY

The histological anatomy with particular reference to musculature, digestive and reproductive organs was worked out in detail. Other structures were not studied in such detail.

### MUSCULATURE

The musculature can broadly be divided into 'somatic or unspecialized musculature' which are arranged in the interchordal zones of the hypodermis and the 'specialized musculature' are those associated with a particular organ.

#### Somatic musculature (Fig. IX, I-K):

The somatic musculature consists of a single layer of cells situated beneath the hypodermis within the interchordal zones. The base of the cells is attached to the hypodermis throughout its length. In each quadrant four cells are present which shows that the nematodes are typically meromyarian. The cells are typically of platymyarian type. The shape and size of the muscle cells may be variable at the same or at different levels of body. Each cell consists of a sarcoplasmic zone containing a nucleus and a fibrillar zone which has ribbon-shaped fibrils.

### Specialized Musculature:

#### (i) Cephalic muscles:

These are two sets of muscles. One set is attached to the lips and is known as labial muscles. The other set is attached to the walls of the stoma and is called stomatal muscles.

##### a. Labial muscles (Fig. IX, B and X, D : 1):

These are six in number bifurcated at their anterior ends. The forked bands of labial muscles are attached to adjoining lips. Thus each lip is bounded by two different labial muscles. Most probably dots near the middle of lips on either side of the internal labial papillae are the points of attachment of these muscles.

##### b. Stomatal muscles (Fig. IX, B and X, D : 2):

These are eight in number attached to the vertical walls of stoma. The point of their attachment is posterior to labial muscles. In cross section of body at level of stoma it becomes evident that two of these are subdorsal, two subventral and four sublateral in position. The contraction and relaxation of these muscles brings about the opening and closing of stoma.

#### (ii) Vulval muscles:

The muscles associated with vulva bring about its dilation and contraction. Basically two sets of muscles are found associated with the vulva.

a. Dilator vulvae (Fig. IX, C&D : 3):

These are 16 in number appearing in 8 pairs, 4 of which are anterior to vulva and 4 posterior. Originating from vulva, they run ventrolaterally and some of them are bifurcated at their ends. The contraction of these muscles opens the vulva.

b. Constrictor vulvae (Fig. IX, D : 4):

The constrictor vulvae are 4 in number and occur in pairs. Each pair originates from base of vagina and is attached to ventrolateral body wall. The muscles function in direction opposite to the dilator muscles and bring about the closure of vulva.

(iii) Sphincter muscles (Fig. IX, A and XI, D : 5):

The oviduct leads to uterus through a valve called as sphincter. The lumen of the valve is controlled by a set of circular muscles which are called as the sphincter muscles.

(iv) Anal muscles (Fig. IX, E & H : 6):

These muscles are associated with the anus. They originate below the anus, run laterally and ventrodorsally. The number of the anal muscle bands is 2 to 3 in females but single in males.

(v) Copulatory muscles (Fig. IX, F & H and XII, F, H & I : 7):

The copulatory muscles extend from the region where the vas deferens and intestine appear constricted and continue

up to the upper margin of cloaca. Each copulatory muscle originates at the laterodorsal body wall, runs lateroventrally and is finally attached between the somatic muscles and ventral chord. The anterior most copulatory muscles are not attached to the dorsolateral body wall but surround the intestine from its dorsal side. In this region the intestine appears transparent. The number of these muscles varies from 30 to 35 on either side of the body wall.

(vi) Accessory copulatory muscles (Fig. IX, F and XII, G : 8):

These are 14 to 16 in number, lying in front of copulatory muscles, of these 8-10 are rather faint, oblique bands followed by 6-8 transversely oriented muscles. The latter run from the dorsolateral body wall of one side to the lateroventral wall of the other side. They cross each other ventral to vas deferens and usually bifurcate before their lateroventral attachment. These muscles along with the first copulatory muscles form a powerful constricting unit around the intestine and vas deferens.

(vii) Gubernacular muscles:

These muscles are associated with gubernaculum and are basically of two types:

a. Retractor gubernaculi (Fig. IX, H and XII, H & I : 9):

They are 4 in number extending from the laterodorsal body wall to the gubernaculum. The anterior most originates



dorsal to the copulatory muscles, curves on the inner side ventral to the spicules and is attached to the anterior end of the gubernaculum ventrally. The last muscle band runs laterodorsally and is attached to the gubernaculum on its laterodorsal side while the rest are attached laterally.

b. Protractor gubernaculi:

The protractor gubernaculi are indistinct.

(viii) Spicular muscles:

These are two sets of muscles attached to the spicules, one retractor spiculi and the other protractor spiculi.

a. Retractor spiculi (Fig. IX, G : 10):

A pair of muscle bands extending from the head of the spicules to the dorsolateral body wall.

b. Protractor spiculi (Fig. IX, G : 11):

A pair of muscle bands surrounding the spicules from dorsal, lateral and lateroventral sides. These are attached to the spicular sheath which in turn envelops the spicules. One muscle band extends posteriorly from the head of the spicules beyond the gubernaculum and connects the dorsal and laterodorsal parts of the gubernaculum, then extends further posteriorly and gets bifurcated. The bifurcated bands are attached to the ventral body wall a short distance behind the cloaca. Another muscle band extends posteriorly from the head of the spicules

runs lateroventrally and then bifurcates. A forked band extends laterally from the spicules and is attached to the laterodorsal parts of it. Another forked band passes laterally from the lateral accessory pieces and is attached with the ventral body wall near the upper lip of cloaca.

(ix) Caudal copulatory muscles (Fig. IX, H : 12):

Posterior to the ~~cloaca~~ there are about 5-9 bands of muscles arranged one behind the other. They extend from the laterodorsal body wall to the ventral body wall. The anterior most muscle band is attached to the lower lip of cloacal opening.

## DIGESTIVE ORGANS

The digestive organs consist of stoma, oesophagus, oesophago-intestinal junction, intestine, rectum and anus. These organs have been described below in detail.

Stoma (Fig. I, C-E and X, A-E):

The stoma opens anteriorly through a hexaradiate oral aperture or vestibulum guarded by six lips. The vestibulum leads to a barrel-shaped buccal cavity formed by two sets: one vertical and another oblique set. Each set has three walls, one dorsal and the other two subventral in position. The dorsal wall of vertical set bears a large dorsal tooth in the posterior half of buccal cavity pointing anteriorly. The subventral vertical walls are provided with denticles opposite the dorsal tooth. The subventral oblique walls bear two foramina each. Though the walls of both sets are almost similar in shape but those of the oblique set are smaller and less sclerotized. A cross section through vertical set shows the three walls to be similar anterior to dorsal tooth. The buccal cavity narrows posteriorly and joins the oesophageal lumen.

A few abnormal types of buccal cavities were also observed. In one specimen the buccal cavity lacked dorsal tooth (Fig. II, A), in other subventral denticles were absent on one of the vertical walls (Fig. II, B). In another case the subventral denticles were

absent from both the walls (Fig. II, C).

Oesophagus (Fig. I, E and X, F-K):

The buccal cavity is followed by a long cylindrical and highly muscular oesophagus. The oesophagus is slightly expanded at its anterior end surrounding the basal part of stoma, but slightly narrowing at level of nerve ring, then widening gradually towards the base. The lumen of the oesophagus is tri[radi]ate, one arm projecting ventrally and the others two subdorsally. Various kinds of thickenings occur on the walls of the lumen. These serve as points of attachment for the radial muscles which originate from the walls of oesophagus. These muscles are strongly developed in the middle of oesophagus. The width of the lumen of oesophagus is uniform throughout its length except at the base of buccal cavity where it is wider.

Oesophago-intestinal junction (Fig. I, E and X, J&K):

The oesophago-intestinal junction or cardia is tuberculate type. It consists of tubercles, a filtration valve and a conical organ (Clark, 1960a). The tubercles are three in number, one of which is situated ventrally and the other two subdorsally. These open into the lumen of oesophagus which in turn opens into a funnel-shaped filtration valve situated in the somatic zone which lies below the transparent zone. The conical organ is heart-shaped projecting into the lumen of intestine. It also has

tri<sup>l</sup>radiate lumen, but lacks cuticularized thickenings.

Intestine (Fig. X, L):

The intestine is a long uniform tube with a wide lumen, narrowing posteriorly in the region where it joins the rectum. In a cross section it is single-layered with 6-8 cells in the circumference. The cells have distinct nucleus but are irregular in shape. The intestine on its dorsal wall where it is surrounded by anterior most copulatory muscles lacks granules and appears hyaline. The lumen of the intestine is also covered by a hyaline layer, but no cilia or rod-like structures are present.

Rectum (Fig. IX, E and X, M):

The rectum is a narrow, dorsoventrally flattened tube, lined with cuticle. In males the rectum along with the genital duct opens into cloaca. The cloaca opens outside through the cloacal aperture.

Anus (Fig. IX, E and X, N):

It is a circular opening placed mid-ventrally.

Rectal glands (Fig. XII, A):

In males four rectal glands are present on each side dorsal and posterior to spicules. Their ducts run a short distance parallel to spicules, turn upwards and open into the cloaca.

## REPRODUCTIVE ORGANS

The sexes are separate. The reproductive organs of the female and male are described below.

### Female reproductive organs (Fig. XI, A-F):

The female gonads are amphidelphic with reflexed ovaries. Each sexual branch consists of an ovary, oviduct, sphincter, uterus, vagina and vulva.

#### Ovary (Fig. XI, A):

The ovary is a thin-walled epithelial sac enclosing the germ cells and lying on ventral side of the body. The epithelium of the sac consists of long flattened cells which are clearly visible towards the proximal end. The germ cells are proliferated at the apical end of the ovary (telogony). Typically the ovary may be divisible into two zones: i) the germinal zone in which rapid division of germ cells takes place, and ii) the growth zone in which a gradual increase in the size of the proliferated cells takes place by gradual accumulation of deutoplasm (Fig. XI, A & B). The oviduct is connected to the ovary subterminally which results in the formation of a blind sac in which the ripening oocytes or oogonia grow until they reach their full length. When the oocytes have reached maturity they leave the ovary and pass into the oviduct.

Oviduct (Fig. XI, A-C):

The ovary is followed by an oviduct which is at first a narrow tube and then enlarged proximally. The distal part differs from the proximal in having transverse markings (Coomans, 1964). In a cross section the distal part appears circular with a narrow lumen and made up of 6-7 high columnar cells. The proximal part also appears cellular in structure but the outlines of the individual cells are indistinguishable from one another and the whole region gives a rather granular vacuolated appearance.

Sphincter (Fig. XI, D):

It is a well developed structure present between oviduct and the uterus. It is a truncated cone-like structure having longitudinal cuticularized linings of fine strands or fibers which arise from the inner walls of the uterus and converge at the centre of sphincter where they become slightly thickened and more refractive in nature. The fibers from the centre form the linings of the proximal end of the oviduct in the lumen of which narrow end of the sphincter is protruded.

Uterus (Fig. XI, E):

The uterus is a highly extensible tube with low columnar epithelial cells in circumference. The distal part of the uterus serves as a fertilization chamber while the eggs are present in the proximal part and are provided with a visible

shell. The maximum number of eggs recorded at one time in the uterus was only 2. The sperms were also seen in the proximal part of uterus of some specimens.

Vagina (Fig. XI, F) :

The uteri of both sides join to form a common short tube, the vagina. It is lined with cuticle and also adequately supported with muscles. It opens outside through vulva.

Vulva (Fig. IX, C & D and XI, F):

It is a ventral transverse slit, post-equatorial in position. It has cuticularized lips formed by the invagination of the cuticle. On either side of the vulva there are present vulval papillae which are variable in number. The prevulval vary from 0-4, and postvulval 0-3. In one female, the cuticularized pieces were absent (Fig. II, H).

Vulval glands (Fig. XI, F):

A pair of vulval glands are situated ventrally which open into the vulva through their ducts.

Male reproductive organs (Fig. XII, A-I):

The male gonad consists of testes, vas deferens, ejaculatory duct and cloaca.

Testes (Fig. XII, A & E):

There are two testes, one directed anteriorly and the other posteriorly. Each testis is enclosed in a thin-walled



sac made up of epithelial cells. Like ovaries these are also telogonic in nature. Each testis is differentiated into a germinal zone and a zone of maturation. The germ cells (spermatogonia) in the germinal zone are smaller in size each with a distinct nucleus. At the distal end of the maturation zone there are usually present several rows of spermatocytes. These are rectangular or hexagonal in shape and give rise to spindle-shaped spermatids which are present in clusters. The length of spermatids may be variable. These ultimately transform into spermatozoa or sperms (Aboul-Eid, 1969). The sperms (Fig. XII, A & C) are numerous and fill the entire proximal end of the growth zone.

Vas deferens (Fig. XII, F & G):

The proximal ends of the two testes join a single long thin-walled tube, the vas deferens. It runs posteriorly up to the constriction formed by the last accessory copulatory muscles and first copulatory muscle. The vas deferens at this point appears very much constricted.

Ejaculatory duct (Fig. XII, A):

Posterior to the horizontal accessory copulatory muscles, the vas deferens differentiates into a thin-walled tube, the ejaculatory duct. It narrows posteriorly to join the rectum to form a cloaca.

### Cloaca and associated structures:

#### Cloaca (Fig. XII, A & I):

The ejaculatory duct narrowing posteriorly joins the rectum to form a common chamber, the cloaca. The middorsal wall of the cloaca is first flattened, then compressed and finally folds backward where the spicules enter the cloaca. A buldging of the midventral wall also appears about halfway its length.

#### Spicules (Fig. XII, D, H & I):

These are a pair, equal in size and similar in shape. They are long and slender ventrally arcuate, sclerotized, narrowing towards their tips. Each spicule may be differentiated into two regions, a head region (capitulum) and a blade (lamina). No region similar to shaft (calmus) was found to be present. Each spicule is covered by a spicular sheath. In a cross section at level of head of spicules, they appear flattened with two cavities and gradually inward curving flanges.

#### Gubernaculum and lateral accessory pieces (Fig. XII, D & H):

The gubernaculum lies dorsal to the spicules from which a pair of lateral accessory pieces extend forward on either side of the spicules. In a cross section the gubernaculum appears trough-shaped with lateral parts first extending along the spicules and lateral accessory pieces (crura) then turned inwards in front of the crura and finally fused with the lateroventral

walls of the cloaca as in Anatonchus amiciae by Coomans & Lima (1965) and Cobbonchus pounamua by Clark (1960~~e~~). The lateral accessory pieces are cuticularized and bifurcated at their distal end.

Supplements (Fig. XII, B):

These are preanal and ventromedian and are connected to the nervous system through a nerve fibre. Each supplement is an elevated structure supplied with a nerve ending.

Ejaculatory glands (Fig. XII, A):

The ejaculatory glands are four in number. They are ovoid or rounded lying over a part of the intestine or ejaculatory duct or both. Posteriorly each one narrows and joins a duct which opens into the lumen of the ejaculatory duct. In one male the ejaculatory glands were five in number (Fig. II, I).

Tail

The tail is long filiform, tapering uniformly, with a ventral curvature and slightly clavate tip. The tail is provided with 4 prominent caudal papillae. The three caudal glands lead to a common duct which opens into an ampulla-like structure, the spinneret. The tuboid opening of the spinneret is provided with a cone-like structure which acts as a valve (Fig. ~~XII~~A and ~~X~~, O).

Apart from the normal type of tails in females, some abnormal tails were also recorded. In one specimen the tail is elongate conoid about half the normal tail length. The caudal glands are situated posteriorly and the spinneret absent (Fig. II, E). In another specimen the tail is provided with a mucro (Fig. II, G). Often the tail is broad especially in the hinder region (Fig. II, F).

## JUVENILE STAGES

First stage Juvenile (Fig. XIII):

Dimensions: Table IV

Description: Body almost straight upon fixation, tapering slightly anteriorly, but markedly posteriorly. Cuticle(inner and outer) smooth, 2 um thick at various places on the body. Lip region marked off, 18-24 um wide, 5-11 um high, distinctly wider than the adjoining body. Amphids small cup-like, apertures 3-4 um wide at 11-12 um from anterior end of body.

Buccal cavity 22-24 x 9-11 um. Apex of dorsal tooth at 5-8 um from base of stoma. The dorsal wall of buccal cavity 18-20 um in length. No tooth or teeth present on the vertical subventral walls. The oblique subventral walls have two foramina on each. Orifices of the oesophageal glands located as follows: dorsal 88-94 um from anterior end of body; the first pair of subventrals 30-39 um from the orifice of dorsal one; the second pair 44-60 um from the orifice of the first pair. Oesophago-intestinal junction tuberculate. Nerve ring at 53-68 um from anterior end of body. Rectum 14-21 um or about one anal body-width long. Genital primordia 30-33 um long. Tail 83-147 um or about 5-7 anal body-widths long. Caudal glands well developed, opening terminal; caudal papillae invisible.

Second Stage Juvenile (Fig. XIV):

Dimensions: Table IV

Description: Body arcuate upon fixation, tapering slightly anteriorly, but markedly posteriorly. Cuticle (inner and outer) smooth, 2 um thick at midbody. Lip region marked off 24-27 um wide, 9-11 um high, distinctly wider than adjoining body. Amphids small cup-like, apertures 3-4 um wide at 11-13 um from anterior end of body.

Buccal cavity 29-31 x 14-17 um. Apex of dorsal tooth at 9-12 um from base of stoma. Dorsal wall 24-28 um high. The vertical subventral walls bear 1-2 teeth and the oblique walls have two foramina on each. Orifices of oesophageal glands located, as follows: dorsal 102-156 um from anterior end of body; the first pair of subventrals 46-53 um from the orifice of dorsal one; the second pair 68-95 um from the orifice of the first pair. Oesophago-intestinal junction tuberculate. Nerve ring at 88-110 um from anterior end of body. Rectum 18-24 um or about 1-2 anal body-widths long. The genital primordia 40-42 um long. Tail elongate-conoid, 141-221 um or about 6-8 anal body-widths long. Caudal glands well developed, opening terminal. Caudal papillae invisible.

Third Stage Juvenile (Fig. XV):

Dimensions: Table IV

Description: Body almost straight upon fixation, tapering slightly anteriorly, but markedly posteriorly. Cuticle (inner and outer) smooth, 2-4 um thick at various places on the body. Lip region marked off, 29-33 um wide, 11-15 um high, distinctly wider than the adjoining body. Amphids small cup-like, apertures 5-6 um wide at 12-15 um from anterior end of body.

Buccal cavity 35-39 x 17-20 um. Apex of dorsal tooth at 13-17 um from base of stoma. The dorsal wall of buccal cavity 27-33 um. Vertical subventral walls bear 3-4 teeth and the oblique walls have two foramina on each. Orifices of oesophageal glands located as follows: dorsal 170-194 um from anterior end of body; the first pair of subventrals 56-63 um from orifice of the dorsal one; the second pair 94-105 um from orifice of first pair. Oesophago-intestinal junction tuberculate. Nerve ring at 113-126 um from anterior end of body. Rectum 23-28 um or about 1-2 anal body-widths long. The genital primordia 57-62 um in length. Tail elongate-conoid, 174-262 um or about 6-7 anal body-widths long. Caudal glands well developed, opening terminal. Caudal papillae 4.

Fourth Stage Juvenile (Fig. XVI):

Dimensions: Table IV

Description: Body almost straight upon fixation, tapering slightly anteriorly, but markedly posteriorly. Cuticle (inner and outer) smooth, 4-6  $\mu$ m thick at various places on the body. Lip region marked off, 35-41  $\mu$ m wide, 12-18  $\mu$ m high, distinctly wider than adjoining body. Amphids small cup-like, apertures 5-6  $\mu$ m wide at 16-18  $\mu$ m from anterior end of body.

Buccal cavity 41-48 x 22-26  $\mu$ m. Apex of dorsal tooth at 17-20  $\mu$ m from base of stoma. The dorsal wall of buccal cavity 37-40  $\mu$ m high. Vertical subventral walls bear 3-6 teeth and the oblique walls have two foramina on each. Orifices of the oesophageal glands are located as follows: dorsal 205-231  $\mu$ m from anterior end of body; first pair of subventrals 65-95  $\mu$ m from orifice of dorsal one; second pair 106-141  $\mu$ m from orifice of first pair. Oesophago-intestinal junction tuberculate. Nerve ring at 120-143  $\mu$ m from anterior end of body. Rectum 25-33  $\mu$ m or about 1-2 anal body-widths long. The genital primordia measures 100-250  $\mu$ m in length. Tail elongate-conoid, 203-314  $\mu$ m or about 6-9 anal body-widths long. Caudal glands well developed, opening terminal. Caudal papillae 4.



## DISCUSSION

The characters for the separation of different juvenile stages conform with those given by Coomans and Lima (1965) with slight deviations. The first stage juveniles differ from all the other stages in the absence of denticles on the vertical subventral walls. These denticles are 1-2 in the second stage, 3-4 in the third stage and 3-6 in the fourth stage juveniles. The genital primordia measure 30-33  $\mu$ m in the first stage, 40-42  $\mu$ m in the second, <sup>stage</sup> 57-62  $\mu$ m in the third stage and 100-250  $\mu$ m in the fourth stage juveniles. In the last juvenile stage usually a rectangular hyaline area also makes its appearance which indicates the place of formation of future vulva.

The presence or absence and number of denticles on the subventral walls are helpful only up to the third stage. The tail length can not be taken <sup>as</sup> a reliable character because it overlaps among the stages. The a, b, c and c' values are not useful distinguishing characters. The other features like amphids, tubercles, spare tooth etc. are also unreliable for the separation of juvenile stages. The presence of spare tooth clearly differentiates the juveniles from the adults.

## VARIABILITY

It is an established fact that morphological characters vary among the individuals of a species, though the degree of variations may differ. The variations may be geographical, ecophenotypic or host induced. Several workers have studied these variations in different species of nematodes (J. B. Goodey, 1952; Rhode and Jenkins, 1957; Bird and Mai, 1965; Fisher, 1965; Bajaj and Jairajpuri, 1977; Geraert, 1978a & b, '79 etc.).

Taylor and Jenkins (1957) reported that deviations from mean value were minimum for vulva position and maximum for tail shape in Pratylenchus spp. Coomans (1962) while studying variations in Rotylenchus goodeyi Loof and Oostenbrink, 1958 found out that b and c values have very wide range of variability. Brzeski (1963) concluded that in Eudorylaimus sp. the value of a has little taxonomic importance while b, c and V are more constant. Sturhan (1963) in his study on Xiphinema and Longidorus species has shown that the body-width, length of oesophagus and tail are not proportional to body length and has concluded that such characters could be useful in these genera. Tarjan (1964) reported differences between two populations of Xiphinema bakeri Williams, 1961 in the length of body, posterior gonad and ratios a, b, c. Bird (1966) found that morphometric and allometric variations among populations of Trichodorus christiei Allen, 1957 are influenced more by host species than their geographical origin. Bird and Mai (1967) confirmed that

in I. christiei Allen, 1957, the length of spear and ratio of V were least variable, while  $G_1$  and  $G_2$  ratios showed maximum variability. According to Thorne and Malek (1968) variations in measurements up to 10% have no diagnostic value at species level. Loof and Maas (1972) while describing intraspecific variations among different populations of Xiphinema spp. concluded that body dimensions alone are unsatisfactory for distinguishing species and qualitative characters should also be given equal importance. Azmi and Jairajpuri (1978) in their studies on Helicotylenchus indicus Siddiqi, 1963 concluded that head height, vulva position, the values of V,  $G_1$  and a in adults; median bulb, excretory pore and values O and c in juveniles are fairly constant characters while other characters show high degree of variations. Morphometric and allometric variations in a population of Xiphinema basiri Siddiqi, 1959 have also been studied by Bajaj and Jairajpuri (1977).

Studies on Hadronchus shakili have shown that there are variations in the specimens, not only from different localities and habitats but also in the specimens from a single population. In the present work, these variations in a single natural population in adults and juveniles of H. shakili collected from soil around roots of Litchi chinensis, Company Gardens, Bareilly, Uttar Pradesh have been statistically analysed.

In the following CV stands for Coefficient of Variability and MSD for Mean  $\pm$  Standard deviation.

## MORPHOMETRIC VARIATIONS

Body length:

The body length varies from 2.40-3.35 mm with MSD =  $2.96 \pm 0.20$  in females, 2.32-3.22 mm with MSD =  $2.82 \pm 0.24$  in males. The CV of this character is 7% in females and 9% in males.

In juveniles the length varies from 0.61-0.99 mm with MSD =  $0.84 \pm 0.12$  and CV = 15% in first stage juveniles; 1.06-1.45 mm with MSD =  $1.26 \pm 0.12$  and CV = 10% in second stage juveniles. In third and fourth stage juveniles, L = 1.49-1.81 mm with MSD =  $1.71 \pm 0.10$ ; 1.81 - 2.31 mm with MSD =  $2.01 \pm 0.12$  respectively. The CV value in both cases is comparatively low being 6% only.

The CV in juveniles as well as in adults shows that total body length is moderately variable except in the first stage juveniles in which it is highly variable (CV = 15%). The total body length among juvenile stages (Fig. XIX) does not show overlapping and thus can be used in the differentiation of juvenile stages.

Body-width:

In females the body-width varies from 61-71  $\mu$ m with MSD =  $66 \pm 2.90$  and CV = 4% while it ranges from 56-62  $\mu$ m with MSD =  $60 \pm 4.94$  and CV = 6% in males.

Among the juveniles, the body-width shows variations from 28-39  $\mu\text{m}$  with  $\text{MSD} = 34 \pm 4.14$  in the first; 35-42  $\mu\text{m}$  with  $\text{MSD} = 40 \pm 2.64$  in the second; 44-53  $\mu\text{m}$  with  $\text{MSD} = 48 \pm 3.44$  in the third and 50-59  $\mu\text{m}$  with  $\text{MSD} = 54 \pm 2.76$  in the fourth stage juveniles. The CV for this character is 12% and 5% in the first and fourth stage juveniles respectively, but is same (7%) for the second and third stage juveniles. The CV values show that body-width is less variable in the females and highly variable in the first stage juveniles.

#### Lip-width:

It varies from 45-60  $\mu\text{m}$  with  $\text{MSD} = 53 \pm 4.89$  and  $\text{CV} = 9\%$  in females; 42-60  $\mu\text{m}$  with  $\text{MSD} = 49 \pm 4.13$  and  $\text{CV} = 8\%$  in males; 18-24  $\mu\text{m}$  with  $\text{MSD} = 21 \pm 1.73$  and  $\text{CV} = 8\%$  in the first stage; 24-27  $\mu\text{m}$  with  $\text{MSD} = 26 \pm 1.0$  and  $\text{CV} = 4\%$  in the second stage; 29-33  $\mu\text{m}$  with  $\text{MSD} = 31 \pm 2.0$  and  $\text{CV} = 6\%$  in the third stage; 35-41  $\mu\text{m}$  with  $\text{MSD} = 38 \pm 2.27$  and  $\text{CV} = 6\%$  in the fourth stage juveniles.

The CV values in adults as well as in juveniles show that this character is moderately variable except in the second stage juveniles where it is less variable. In the third and fourth stage juveniles the extent of variation is same ( $\text{CV} = 6\%$ ).

#### Lip height:

The lip height varies from 15-25  $\mu\text{m}$  with  $\text{MSD} = 18 \pm 1.36$  in females; 15-19  $\mu\text{m}$  with  $\text{MSD} = 17 \pm 0.13$  in males. The CV for

this character is 8% and 7% in females and males respectively.

Among the juveniles, the lip height measures 5-11 um with  $MSD = 8 \pm 1.08$  and  $CV = 14\%$ ; 9-11 um with  $MSD = 10 \pm 1.19$  and  $CV = 12\%$ ; 11-15 um with  $MSD = 13 \pm 1.26$  and  $CV = 10\%$ ; and 12-18 um with  $MSD = 16 \pm 2.17$  and  $CV = 14\%$  in the first, second, third and fourth stage juveniles respectively.

In the juveniles this character appears to be highly variable, maximum being 14% in the first and fourth stage juveniles while in adults its variation being moderate (7-8%). Thus the extent of variation for the lip-width as well as lip height is same in the adults while in the juveniles the lip height is more variable than the lip-width.

#### Width of amphid apertures:

The width of amphid apertures, ranges from 5-8 um with  $MSD = 7 \pm 0.50$  in females, and 6-7 um with  $MSD = 6.7 \pm 0.25$  in males. The CV for this character is 7% and 4% in females and males respectively.

The CV for this character in juveniles is 7-10%. It measures 3-4 um with  $MSD = 3.8 \pm 0.40$  and  $CV = 10\%$  in the first stage; 3-4 um with  $MSD = 3.9 \pm 0.39$  and  $CV = 10\%$  in the second stage; 5-6 um with  $MSD = 5.6 \pm 0.40$  and  $CV = 7\%$  in the third stage and 5-6 um with  $MSD = 5.8 \pm 0.40$  and  $CV = 7\%$  in the fourth stage juveniles.

The CV of this character shows that among all the juvenile stages and adults it is moderately variable.

Position of amphid apertures from anterior extremity:

The position of amphid apertures is highly variable in both the sexes, with CV being 14-16%. It ranges from 15-23 um with MSD =  $18 \pm 2.54$  in females; and 14-22 um with MSD =  $18 \pm 2.88$  in males.

In the first stage juveniles it is 11-12 um with MSD =  $11.5 \pm 0.50$  and CV = 4%; in the second stage 11-13 um with MSD =  $12 \pm 0.67$  and CV = 6%; in the third stage; 12-15 um with MSD =  $14 \pm 0.90$  and CV = 6%; and in the fourth stage juveniles 16-18 um with MSD =  $17 \pm 0.91$  and CV = 5%.

Buccal cavity:

Of all the morphometric characters studied in the adults the length of buccal cavity is least variable. It measures 50-65 um with MSD =  $60 \pm 0.71$  and CV = 2% in females, and 49-60 um with MSD =  $55 \pm 1.63$  and CV = 3% in males.

The CV values for this character in juveniles are 3% for the first stage; 4% for the second stage; and 5% for the third and fourth stages. It measures 22-24 um with MSD =  $23 \pm 0.62$  in the first; 29-31 um with MSD =  $30 \pm 1.05$  in the second; 35-39 um with MSD =  $37 \pm 1.76$  in the third; and 41-48 um with MSD =  $45 \pm 2.23$  in the fourth stage juveniles.

The low CV values in adults as well as in the juveniles indicate that the length of buccal cavity is a fairly constant character. It is also evident that length of buccal cavity does not show overlapping in the various juvenile stages (XIX, A) and hence it can be used to differentiate the various juvenile stages.

The width of buccal cavity ranges from 32-40  $\mu$ m with MSD =  $36 \pm 1.31$ , and 29-35  $\mu$ m with MSD =  $31 \pm 1.34$  in females and males respectively. The value of CV in both the sexes for this character is only 4%. In the first, second, third and the fourth stage juveniles it measures 9-11  $\mu$ m with MSD =  $10 \pm 0.62$  and CV = 6%; 14-16  $\mu$ m with MSD =  $14 \pm 0.76$  and CV = 5%; 17-20  $\mu$ m with MSD =  $19 \pm 1.18$  and CV = 6% and 22-26  $\mu$ m with MSD =  $25 \pm 1.55$  and CV = 6% respectively. The low values of CV show that the width of buccal cavity is less variable except in the third and fourth stage juveniles (CV = 6%). Like the length, the width of buccal cavity also does not show overlapping in the different juvenile stages and hence is useful in their separation.

#### Dorsal tooth:

The position of dorsal tooth from base of buccal cavity varies considerably in both the sexes, with CV being equal to 10% in males and 8% in females. It ranges from 21-30  $\mu$ m with MSD =  $26 \pm 2.20$  in females, and 19-27  $\mu$ m with MSD =  $23 \pm 2.31$  in males.



In the juveniles the CV of dorsal tooth is 15%, 11%, 3% and 5% in the first, second, third and the fourth stage juveniles respectively. It measures 5-8 um with MSD =  $7 \pm 1.02$  in the first stage; 9-12 um with MSD =  $10 \pm 1.10$  in the second stage; 13-17 um with MSD =  $15 \pm 1.26$  in the third stage; and 17-20 um with MSD =  $18 \pm 0.86$  in the fourth stage juveniles.

The CV values of this character in various developmental stages show that it is highly variable in the first and second stage juveniles, moderately in the adults and less variable in the third and fourth stage juveniles.

#### Oesophageal length:

The length of oesophagus measures 476-716 um with MSD =  $662 \pm 42.33$  and CV = 6% in females, and 500-685 um with MSD =  $629 \pm 48.42$  and CV = 8% in males. This indicates that in the adults this character is moderately variable.

In the first stage juveniles, the oesophageal length varies from 188-280 um with MSD =  $249 \pm 26.78\%$  and CV = 11%. In the second stage it measures 294-362 um with MSD =  $328 \pm 22.12$  and CV = 7%. In the third and fourth stage juveniles, the oesophageal length shows a lesser degree of variability and the CV = 3% and 4% respectively. It ranges from 378-430 um with MSD =  $409 \pm 17.10$  in the third, and 452-512 um with MSD =  $479 \pm 13.13$  in the fourth stage juveniles. It is evident from the present study that the oesophageal length exhibits more

variations in the first stage than in the other stages. This study also shows that the length of oesophagus shows no overlapping in the juvenile stages (Fig. XIX<sup>6</sup>).

#### Nerve ring:

The position of nerve ring from anterior extremity is less variable in the adults with CV being 6% in females and 5% in males. It measures 150-198 um with MSD =  $176 \pm 10.63$ ; 145-178 um with MSD =  $171 \pm 7.98$  in females and males respectively.

In the juveniles, the position of nerve ring shows a lesser degree of variability and the CV is 2%, 3%, 2%, 4% in the first, second, third and fourth stage juveniles respectively. It measures 53-68 um with MSD =  $64 \pm 1.95$ ; 88-110 um with MSD =  $102 \pm 3.26$ ; 113-126 um with MSD =  $124 \pm 2.42$  and 120-143 um with MSD =  $133 \pm 5.06$  in the first, second, third and fourth stage juveniles respectively.

#### Vagina:

In females the length of vagina varies from 19-25 um with MSD =  $23 \pm 2.01$ . The CV for this character is 9% indicating moderate variation.

#### Vulva:

The position of vulva from anterior end varies moderately with CV being 8%. The distance being 1490-2140 um with MSD =  $1821 \pm 143$ .

Gonads:

The lengths of anterior and posterior gonads are highly variable, CV being 16% and 12% respectively. The anterior gonad measures 272-488 um with MSD =  $366 \pm 58.83$ ; and posterior gonad 260-450 um with MSD =  $360 \pm 43.63$  respectively.

Spicules:

The length of spicules varies from 91-106 um with MSD =  $96 \pm 4.78$ . The coefficient of variability for this character is only 5% which clearly shows that this character is less variable and can be used safely at specific level.

Gubernaculum:

The length of gubernaculum ranges from 18-24 um with MSD =  $21 \pm 2.04$  and CV = 10%. This indicates that the length of gubernaculum is moderately variable.

The length of lateral accessory pieces varies from 12-18 um with MSD =  $13 \pm 1.54$  and CV = 12%. This high value of CV clearly indicates that the length of lateral accessory pieces is highly variable.

Ventromedian supplements:

The number of ventromedian supplements varies from 11-16 with MSD =  $13 \pm 1.84$ . The CV value is 14% showing thereby that the number of ventromedian supplements is highly variable.

Rectum:

The CV value of length of rectum in females is 11% and it measures 31-52  $\mu\text{m}$  with  $\text{MSD} = 41 \pm 5.19 \mu\text{m}$ .

In juveniles it varies from 14-21  $\mu\text{m}$  with  $\text{MSD} = 17 \pm 1.37$  and  $\text{CV} = 8\%$  in the first stage; 18-24 <sup>$\mu\text{m}$</sup>  with  $\text{MSD} = 21 \pm 1.21$  and  $\text{CV} = 6\%$  in the second stage; 23-28  $\mu\text{m}$  with  $\text{MSD} = 26 \pm 1.48$  and  $\text{CV} = 6\%$  in the third stage; and 25-33  $\mu\text{m}$  with  $\text{MSD} = 27 \pm 1.55$  and  $\text{CV} = 6\%$  in the fourth stage juveniles.

The value of CV in the adults and juveniles shows that the length of rectum in the adult is highly variable but in the juveniles its variation is moderate.

Anal body-width:

In females the anal body-width varies from 41-53  $\mu\text{m}$  with  $\text{MSD} = 47 \pm 2.95$  and  $\text{CV} = 6\%$  while in the males it measures 47-55  $\mu\text{m}$  with  $\text{MSD} = 53 \pm 2.42$  and  $\text{CV} = 5\%$ .

In juveniles the anal body-width varies from 18-24  $\mu\text{m}$  with  $\text{MSD} = 21 \pm 2.79$  and  $\text{CV} = 13\%$  in the first stage; 24-30  $\mu\text{m}$  with  $\text{MSD} = 28 \pm 1.96$  and  $\text{CV} = 7\%$  in the second stage; 31-36  $\mu\text{m}$  with  $\text{MSD} = 34 \pm 1.92$  and  $\text{CV} = 6\%$  in the third stage and 34-39  $\mu\text{m}$  with  $\text{MSD} = 36 \pm 1.73$  and  $\text{CV} = 5\%$  in the fourth stage.

It is evident from CV values that this character has a lesser degree of variability in adults as well as in the juveniles with the exception of first stage juvenile which exhibits high

degree of variation (13%).

#### Tail length:

The variability of same extent in length of tail has been noted in both sexes as the CV being 13% and 14% in males and females respectively. In females the length of tail varies from 262-430  $\mu\text{m}$  with  $\text{MSD} = 372 \pm 47.72$  and in males 245-335  $\mu\text{m}$  with  $\text{MSD} = 297 \pm 41.78$ .

In juveniles the tail measures 83-147  $\mu\text{m}$  with  $\text{MSD} = 123 \pm 27.42$  and  $\text{CV} = 22\%$  in the first; 141-221  $\mu\text{m}$  with  $\text{MSD} = 175 \pm 17.53$  and  $\text{CV} = 10\%$  in the second; 174-262  $\mu\text{m}$  with  $\text{MSD} = 213 \pm 18.40$  and  $\text{CV} = 9\%$  in the third; and 203-314  $\mu\text{m}$  with  $\text{MSD} = 246 \pm 28.05$  and  $\text{CV} = 11\%$  in the fourth stage juveniles.

The high value of CV in adults and juveniles clearly shows that tail length is greatly variable, maximum variation being in the first stage juveniles. Since there is an overlapping among the successive stages, this character can not be utilized in differentiation of various juvenile stages (Fig. XIX, C).

### ALLOMETRIC VARIATIONS

#### Body length/body-width (a):

The value of a varies from 35-52 with  $\text{MSD} = 45 \pm 4.09$  and  $\text{CV} = 9\%$  in females and 35-54 with  $\text{MSD} = 47 \pm 3.87$  and  $\text{CV} = 8\%$  in males. In juveniles the CV for this character is 11%,

6%, 5% and 6% in the first, second, third and the fourth stage juveniles respectively. The value of  $a$  varies from 21-31 with  $MSD = 26 \pm 2.84$  in the first stage, 29-35 with  $MSD = 32 \pm 1.93$  in the second stage; 33-38 with  $MSD = 35 \pm 1.87$  in the third stage and 34-43 with  $MSD = 37 \pm 2.21$  in the fourth stage juveniles.

The moderate values of CV in juvenile as well as in the adults show that it is moderately variable except in the first stage juveniles in which it is highly variable. The present study reveals that the body-width in different developmental stages increases with the increase in body length but not in the same proportion. The study further indicates that  $a$  and body length ( $L$ ) bear equal taxonomic weight.

#### Body length/oesophageal length ( $b$ ):

The value  $b$  varies from 3.8-6.2 with  $MSD = 4.45 \pm 0.37$  and  $CV = 8\%$  in females and 4.2-5.2 with  $MSD = 4.45 \pm 0.40$  and  $CV = 9\%$  in males. Since the CV of  $b$  is greater than CV of oesophageal length (6% in females and 8% in males), it is evident that  $b$  is more variable than the oesophageal length. In juveniles, the CV for this ratio is 9% and 8% in the first and second stage, 3% in the third and fourth stage juveniles. It ranges 2.5-3.7 with  $MSD = 3.3 \pm 0.31$ ; 3.4-4.5 with  $MSD = 3.8 \pm 0.30$ ; 3.9-4.2 with  $MSD = 4.1 \pm 0.11$  and 4.0-4.5 with  $MSD = 4.1 \pm 0.14$  in the first, second, third and the fourth stage

juveniles respectively.

Fig. XVII, B and XIX, B show that oesophageal length does not depend entirely upon the body length. The largest specimens have same oesophageal length as the shortest ones or vice versa.

Body length/tail length (c):

The tail length is negatively correlated with the total body length both in adults as well as in the juveniles (Fig. XVII, C and XIX, C), thus making the value of c much variable. The CV for this value in males and females is 9% and 10% respectively. It ranges from 7.3-11.8 with  $MSD = 9.2 \pm 0.80$  and 6.4-11.8 with  $MSD = 8.0 \pm 0.81$  in males and females respectively. In juveniles it is 5.5-7.9 with  $MSD = 6.7 \pm 0.63$  and  $CV = 9\%$  in the first; 6.3-7.7 with  $MSD = 7.1 \pm 0.49$  and  $CV = 7\%$  in the second; 7.4-8.6 with  $MSD = 8.0 \pm 0.43$  and  $CV = 5\%$  in the third; and 6.6-9.1 with  $MSD = 8.0 \pm 0.75$  and  $CV = 9\%$  in the fourth stage juveniles.

Thus the value of c for all the stages is moderately variable and shows overlapping between successive stages.

Body length/buccal cavity length:

The buccal cavity length is independent of total body length in adults as well as in the juveniles (Fig. XVII, A and XIX, A).

Body length/vulva position (V):

The vulva position is positively correlated with the total body length (Fig. XVIII, A) thus making V a stable character. Its value varies from 60-66 with MSD =  $61 \pm 1.62$  and CV = 3%.

Body length/gonad length ( $G_1$  &  $G_2$ ):

The CV for  $G_1$  and  $G_2$  is same (17%). It ranges from 9-15 with MSD =  $12 \pm 1.98$ , and 9-15 with MSD =  $12 \pm 1.42$  in  $G_1$  and  $G_2$  respectively. From the CV value it is evident that  $G_1$  and  $G_2$  have variability greater than all the other characters. From Fig. XVIII, B it is also clear that both the anterior and posterior gonads are negatively correlated with the total body length.

Body length/testes length (T):

The value T varies from 39-45 with MSD =  $43 \pm 2.93$ . The CV for this character is 7% indicating that it is a moderately variable character.

## DISCUSSION

The present morphometric studies on adults and juveniles of Hadronchus shakili indicate that there is variation in all the morphological characters to a certain degree. The morphometric characters evaluated in the adults show that the length and width



of buccal cavity are least variable (CV being 2 to 3% only) and can be regarded as good diagnostic characters at species level. There is considerable variation in total body length, lip-width and lip height, width of amphid apertures, position of dorsal tooth, length of oesophagus and length of gubernaculum. The position of nerve ring, anal body-width and the length of spicules also show a lesser degree of variation but all the other characters are highly variable. In the allometric characters the value of V is least variable. This finding agrees with the observations made by Bird and Mai (1967) on Trichodorus christiei; Tarjan (1969) on Xiphinema americanum; Wu (1960) on Ditylenchus destructor; Rashid and Khan (1978) on Pratylenchus coffeae; and Bajaj and Jairajpuri (1977) on Xiphinema basiri. The  $G_1$  and  $G_2$  values show highest coefficient of variability which partly agrees with Azmi and Jairajpuri (1978). The body width, oesophageal length, tail length and length of gonads are independent of body length. These findings are in conformity with those of Sturhan (1963), Geraert (1968).

The length of buccal cavity is also independent of body length. The extent of variability for values of a and L is almost same while for the value c it shows more variability than the anal body-width. The length of oesophagus is less variable than b. This is in accordance with the findings of Geraert (1968). The value c is more constant than the tail

length.

In juveniles, the first stage exhibits the maximum variability in all morphometric and allometric characters. In third and fourth stage juveniles the degree of variation for each character shows consistency. The results further show that in the juvenile stages and the adults the extent of variation for a particular character is almost same. This indicates that variability of a character is determined from the very first stage (Bajaj and Jairajpuri, 1977).

From the above results Fig. XX, A & B it is also evident that a character having larger standard deviation may have lesser CV value. These observations further support Bird & Mai (1967) suggestion that in taxonomy coefficient of variation should be included for each character in addition to its standard deviation.

## COMPARATIVE DEVELOPMENT

The dimensions of the body of a nematode species though important for distinguishing its various stages are alone not sufficient because these may be influenced by environmental factors (Calpham, 1930; Bingefors, 1957 and van Weerdt, 1958). The development of gonads and other organs, which either grow or are replaced at each moult provide reliable criteria for distinguishing various juvenile stages and the adults as shown by Raski (1950); van Gundy (1958); Yuksel (1960); Chuang (1962); Coomans and Lima (1965); Yuen (1965); Hirschmann and Triantaphyllou (1967); Roman and Hirschmann (1969); Dasgupta et al. (1970); Chin (1977) etc. Mulvey (1961) gave a brief account of the development of buccal cavity and tail of the juveniles and adults of Anatonchus tridentatus (de Man, 1876) De Coninck, 1939. Grootaert and Maertens (1976) also discussed about the moulting and the buccal cavity formation in Mononchus aquaticus Coetzee, 1968.

van Weerdt (1960) studied the development of gonads in Radopholus similis (Cobb, 1893) Thorne, 1949 and concluded that the genital primordium of all nematodes has two germinal nuclei irrespective of the number of gonads in the adults. This supported the views held by Chitwood and Chitwood (1950). Hirschmann (1962) in Ditylenchus trifomis Hirschmann and

Sasser, 1955; Anderson and Darling (1964) in Ditylenchus destructor Thorne, 1945; Ahmad and Jairajpuri (1979) in Chiloplacus symmetricus (Thorne, 1925) Thorne, 1930 have shown that instead of two germinal nuclei there may be only one germinal nucleus in the primordium. Hirschmann (1962) had, however, stated that the number of germinal nuclei in the primordium is independent of number of gonads. Pai (1928) was the first to report and distinguish the germinal nuclei from the somatic ones. He showed that in Turbatrix aceti (Müller, 1783) Peters, 1927, the genital primordium in the first stage juvenile consists of two germinal nuclei and two somatic nuclei. Usually the sexual differences become evident at the third juvenile stage in most species (van Gundy, 1958; Triantaphyllou and Hirschmann, 1960; Anderson and Darling, 1964). However, in some nematodes it may be evident even at the second stage as in species of Heterodera and Meloidogyne. The development of the genital tract and ovary in different seasons of the year was studied by some workers, e.g., Griffin and Darling, 1964; Flegg, 1967; Jairajpuri and Bajaj, 1978 etc.

In the present work, an attempt has been made to study the development of various organs in Hadronchus shakili with special reference to its buccal cavity, oesophagus, gonads, tail etc. and to use these for distinguishing the sexes and the juvenile stages. Observations were made on moulting stages and the development of genital tract and the ovary in different seasons.

The observations on the comparative development of the buccal cavity and other organs were based on specimens including moulting stages from a natural population obtained from Bareilly, Uttar Pradesh. For the study of development of gonads, live nematodes were stained in 1% acetic orcein and observed in a drop of dilute stain. In order to study the seasonal development of the genital tract specimens were collected and studied at monthly intervals from July 1977 through June 1978.

#### OBSERVATIONS

##### Buccal cavity (Fig. XXI, A-C):

The buccal cavity was formed anew at each moulting stage and reached its full size before the initiation of next moult. The comparative measurements of the buccal cavities of the juveniles, moulting stages and the adults are given in Table V.

As the moulting initiated, the lip region appeared slightly swollen and straight with the development of a new cuticle underneath it. Later, the old and the new cuticles separated clearly and a distinct space appeared in between (Fig. XXI, A). In the early stages of moulting the walls of the new buccal cavity made their appearance in the form of thin membranous plates just outside the old buccal cavity. The vertical walls of the new buccal cavity were laid down very close

to vertical walls of the old one but the gap was a bit wider towards the base. From the very beginning the oblique walls appeared more prominent than the vertical walls. As development proceeded, these gradually thickened and became more refractive in nature. The oblique walls were first almost similar to each other in shape and size, but differentiated in later stages. The dorsal oblique wall appeared comparatively shorter than the subventrals. The oblique subventral walls increased mainly in length. During each juvenile stage the dorsal tooth appeared hollow with a spare tooth near its base (Fig. XXII). With the increase in the size of buccal cavity the dorsal tooth moved posteriad towards the base of buccal cavity and finally attached itself with the new vertical wall (Fig. XXI, A & B). Now the spare tooth which first appeared like a small conical piece came into the existence (Fig. XXI, C). Thus in the late phase of each moulting stage there might be three dorsal teeth. The first of these was the tooth of the previous juvenile stage which was in the process of being cast off along with the old buccal cavity, the second one was the functional tooth of that particular stage and the third one being the new spare tooth which would become the functional tooth of the future stage (Fig. XXIII). The dorsal tooth may be hollow in young adults but later it became solid. The subventral teeth and the foramina developed in the later phase of each moulting stage. The subventral teeth were not present in the first stage juveniles,

but in other stages they were always present.

A comparative study of the buccal cavity showed that it gradually increased from the first stage juveniles to the adult, but it developed more markedly in the adults than in the juveniles. The development in the first and second stage was minimum. It was much more pronounced in the adult females than the males. The position of dorsal tooth from the base of buccal cavity when calculated as percentage in relation to the length of buccal cavity was found to be comparatively more in the adults than in the juveniles. Among juveniles, it was noted maximum in the first stage and minimum in the fourth stage. In the second and third stage juveniles it was found to be same.

#### Oesophagus:

At the time of formation of new buccal cavity a new oesophageal linings and new set of tubercles also developed, while the old ones were pulled anteriorly (Fig. XXI, E). At the beginning of each moult, the oesophago-intestinal junction became flattened. Sometimes, the development of new tubercles took place quite late while the old tubercles had already migrated upwards and it appeared as if the oesophago-intestinal junction was a non-tuberculate type (Fig. XXI, D). The oesophageal length in relation to body length was comparatively more in the juveniles than the adults. Among the juveniles, it was maximum for the first stage but was same for the third and fourth stages.

Gonads:First stage juvenile (Fig. XXIV, A):

The primordia in the first stage juvenile consisted of two oval bodies connected by a cellular strand, lying at about 61% from anterior end of body. As will be shown later, these anterior and posterior primordium gave rise to the anterior and posterior gonads respectively. Each oval body contained a single large centrally located spherical nucleus and two somatic nuclei. The two types of nuclei could be differentiated from each other by their taking up stain differently. The germinal nuclei appeared coarse granular with a few dark areas, while the somatic nuclei stained darkly. The nuclei of ventral chord which showed a granular pattern numbered about 23 from middle of primordia to base of oesophagus. Total length of primordia varied from 30-33  $\mu\text{m}$ . During the first moult no obvious change occurred in the primordia.

Second stage juvenile (Fig. XXIV, B and XXV, A):

During this stage, the number of germinal and somatic nuclei remained same but the length of primordia increased to about 42  $\mu\text{m}$ . In some juveniles 2-3 oval nuclei that stained darker than the ventral chord nuclei were also present near the posterior end of primordia. These were the specialized ventral chord nuclei, supposed to be derived from the ventral chord nuclei (Hirschmann, 1962). These nuclei were present only in



those juveniles that were to develop into the females. Thus male and female juveniles could be distinguished during the second stage. The number of ventral chord nuclei from base of oesophagus to the primordia varied from 33-36.

During the second moult (second stage moulting into third stage) the germinal nucleus remained undivided but the somatic nuclei divided giving rise to 4-6 nuclei one of which became the cap nucleus at the distal end and the other at the proximal end of the anterior and posterior primordium respectively (Fig. XXIV, C and XXV, B). These two cap nuclei did not divide any further during the gonad development. The remaining somatic nuclei formed the gonoduct and the epithelial layer of the ovaries and testes during the future course of development of the females and males respectively. The number of ventral chord nuclei remained same as in the second stage. The specialized ventral chord nuclei were four in number (Fig. XXIV, C).

Third stage juvenile (Fig. XXIV, D and XXV, C):

During the third juvenile stage the genital primordia enlarged further. It measured about 62  $\mu$ m in its early stage of development. There were seven nuclei, of which one was the germinal nucleus and the other six somatic nuclei. The latter were positioned as follows: one anterior, one posterior, two dorsal one ventral and one adjacent to the germinal nucleus in each primordium. The somatic nuclei appeared to be migrating towards

the middle of primordia. In male juveniles, a small compact mass of dark staining nuclei also appeared in the anal region. This mass represented the primordia of the future spicule and gubernaculum (Fig. XXV, H). The multiplication of the ventral chord nuclei also took place and these now numbered 54 and spreading from the middle of the primordia to the base of oesophagus.

At the time of the third moulting the primordia further developed and enlarged in both the sexes. The somatic nuclei proliferated further and their number increased in the middle region of the primordia. The most marked developmental change was the division of the germinal nuclei which had remained undivided so far. There were repeated divisions giving rise to 6-8 germinal nuclei in females (Fig. XXIV, E) and 9-11 in males (Fig. XXV, D). At the end of this moult, besides the cap nucleus 3-4 small, darkly stained epidermal nuclei were present in each primordium. The number of the ventral chord nuclei from base of oesophagus to the middle of primordia was avg. 136. In females, the specialized ventral chord nuclei doubled in number and apparently became grouped in fours. In males, the spicular and gubernacular primordia enlarged further by repeated multiplication.

Fourth stage juvenile (Fig. XXIV, F and XXV, E & F):

During this stage there was considerable elongation in the length of the developing gonads. Though both the primordia were

equally developed in the early stages, their further development in females was asymmetrical. The anterior primordium, grew more quickly than the posterior one. In males the development of the gonad continued symmetrically and faster than the females. The germinal nuclei at the end ranged from 11-25 in males and 9-10 in females. The somatic nuclei proliferated further and arranged themselves irregularly along the longitudinal axis of body. The specialized ventral chord nuclei now numbered 16 in the vaginal area with their more inward migration. In males, the spicular and gubernacular primordia advanced further (Fig. XXV, I).

During the fourth moult the gonad development was very rapid. In the early stages the gonads in the female juveniles consisted of reflexed ovaries connected by a long genital duct with a large number of somatic nuclei arranged in an irregular manner. The sphincter which differentiated the genital duct into oviduct and uteri appeared as a small structure with spindle-shaped nuclei and poorly developed cuticular linings. The uteri consisted of a large number of somatic nuclei, while the oviduct had fewer nuclei. In the vaginal area there were 20-24 specialized ventral chord nuclei arranged somewhat in a circular manner with a space in between. With the completion of this moulting, the cuticular linings of the vagina were formed (Fig. XXIV, G & H). In adult females the size as well as the number of germinal nuclei

(oogonia) increased.

In male juveniles the gonad differentiated into testes and gonoduct. The somatic nuclei proliferated and arranged along the gonoduct. In the anterior part of gonoduct (vas deferens) these nuclei were arranged in two rows while in the posterior region (ejaculatory duct) their arrangement was irregular. The germinal nuclei in the testes also undergo further division. Spicules appeared as faint refractory lines during the early stages of the moult but gradually thickened. Simultaneously with the formation of spicules the gubernaculum and the lateral accessory pieces were also formed. The development of the copulatory muscles also took place along with the formation of spicules, but the development of the ventromedian supplements began rather late. From cloaca to the anterior region of ejaculatory duct where the accessory copulatory muscles originated a large number of nuclei running ventrodorsally could also be distinguished. The nuclei close to ventral chord nuclei were difficult to be distinguished but the other type of nuclei stained comparatively darker<sup>and</sup> were more or less spindle-shaped. Moreover, in the region where the accessory copulatory muscles originated, there was accumulation of these nuclei in a triangular area (Fig. XXV, G). In adults, the number of all types of nuclei increased.

Development of genital tract (Fig. XXVI):

The ovaries as well as the total length of the genital tract was much longer in the breeding season (March to October) than during the non-breeding season. The maximum lengths were measured in the month of July for both the ovaries. Afterwards, their lengths decreased gradually till October. From November till February (non-breeding season) there was a sharp reduction in the length of anterior ovary to about 1/4th of its maximum length in February. From March onwards, there was continuous increase in the length till July. The pattern for posterior ovary was almost same except that the reduction was about 1/5th of its maximum length. The length of the anterior ovary was greater than the posterior one in all the months (Fig. XXVI, A). Apart from this, the hind part of the ovaries appeared brownish in colour during the breeding season due to the presence of ripe oocytes which had granular cytoplasm. During the non-breeding season (November-February) the ovaries appeared colourless and contained oocytes with non-granular cytoplasm.

The anterior genital tract very much like the anterior ovary also measured maximum in length in the month of July. Afterwards, it started to reduce till the month of January when it was minimum. From February onwards it again increased in similar fashion as the anterior ovary. The posterior genital tract followed a slightly different pattern. Its minimum length

was reached in the month of November. From December till June there was gradual increase in its length but later on the growth was irregular. The length was almost same in the months of July, October and April. Throughout the year the anterior genital tract was always longer than the posterior genital tract except in the month of January when both were almost equal (Fig. XXVI, B). Immediately before the beginning of the breeding season the intestinal region showed changes similar to those described for species of Xiphinema (Griffin and Darling, 1964; Jairajpuri and Bajaj, 1978).

A large number of gravid females (382) were observed either live or dead and were grouped under three categories: i) those having eggs in the anterior uterus, ii) Eggs in the posterior uterus, and iii) Eggs in both the uteri. The frequency of first category was 58% while of the other two was 21%. From these it can be inferred that there was only 21% chance of both the ovaries producing eggs simultaneously, otherwise only one of the two ovaries was producing eggs at a time. Upon counting the number of eggs in both the branches separately, it was also noted that the frequency of egg development by the anterior branch was much higher (65%) than the posterior branch (35%). This shows that the anterior ovary is much more active in egg production than the posterior one.

Tail (Fig. XXIII, E-H):

With the formation of new cuticle in the anterior region the new cuticle of the tail and lumen of rectum were also laid down simultaneously (Fig. XXI, G). The tail length in relation to body length followed the same pattern as the oesophagus, but in the males it was smaller than in the females. The anal body-width in relation to tail length on the average was more in the males than juveniles, but in the females it was just the reverse. Among juveniles, the development was maximum in the first stage while minimum in the fourth stage juveniles.

#### DISCUSSION

The above observations show that the formation of new cuticle precedes the formation of buccal cavity and becomes fully formed before the old cuticle is cast off. Along with this, the spare tooth of the previous stage also becomes fully developed and a new spare tooth is formed at base of functional tooth. These observations are in agreement with those of Grootaert and Maertens (1976) on Mononchus aquaticus. The last phase of moulting commences with the separation of old and new cuticle by large spaces. First the rupture of the old cuticle takes place at the anterior end. The buccal cavity along with the old oesophageal linings and tubercles is then ejected out. Later, the old cuticle of the tail and rectum becomes loose and soon the

juveniles escape from the exuvium. The other parts which are formed a new are spinneret, amphids, denticles, foramina etc. During the entire development there are four moulting stages (Fig. XXIII). The size of the buccal cavity in different developmental stages shows that the development is minimum in the first and second stages. This might be due to the shorter durations of these stages. Further, the development of oesophagus and tail (in relation to body length) shows that their development is comparatively more in juveniles than the adults.

The primordia in the first stage consists of two oval bodies, each with one germinal nucleus and two somatic nuclei. The somatic nuclei divide for the first time in the second stage while the germinal nuclei in the third moulting stage. As shown by earlier workers (Maupas, 1899, Pai, 1928; Hirschmann, 1962 etc.) the cap nucleus is derived from the somatic nuclei and do not divide later but remains a part of the ovaries or testes. The sexes become differentiated at the second juvenile stage because of the presence of specialized ventral chord nuclei in the females and spicular and gubernacular primordia in the third and fourth stages in the male juveniles. This is quite similar to that reported for D. triformis (Hirschmann, 1962). The development of the gonads in the last stage is comparatively more in the male juveniles than in the females. Moreover, both the gonads are almost equally developed in the male juveniles, whereas in the females one of them grows quicker than the other. These



observations agree with those of Coomans and Lima (1965) on Anatonchus amiciae. The nuclei of the sphincter are also probably derived from the somatic nuclei. The development of the vagina takes place from the specialized ventral chord nuclei. The measurements show that the size of the genital primordia, similar to that of buccal cavity, do not overlap in the different juvenile stages. Hence, it becomes evident that there is some relation although indirectly between the development of gonads and that of the buccal cavity.

The changes in the genital tract during the breeding and non-breeding season agree fully with those of Jairajpuri and Bajaj (1978) for Xiphinema basiri and partially with Flegg (1967) for Xiphinema vuittenezi.

### POPULATION DYNAMICS

The population studies of plant-parasitic and the predatory nematodes are important, since the former is related to the incidence and intensity of crop injuries while the latter acts as an agent of biological control of the plant-parasitic nematodes. The population of a nematode, in a field shows wide fluctuations throughout the year and is dependent upon various biotic and abiotic factors. Although considerable work has been done during the last few decades on the population dynamics of plant-parasitic nematodes (Hollis and Fielding, 1955; Rigs et al., 1956; Wallace, 1962, '63; Norton, 1963; Zuckerman et al., 1964; Szczygiel, 1966; Flegg, 1968; Singh and Misra, 1968; Khan et al., 1971; Szczygiel and Hasiior, 1972a & b; Biro, 1973 etc.), but almost no work has been done on the population dynamics of Mononchida.

The data on the vertical distribution of nematodes obtained by different authors are similar for some species but differ in others. The nematodes, Tylenchorhynchus dubius (Butschli, 1873) Filipjev, 1936, Rotylenchus fallorobustus Sher, 1965 and Tylenchus spp. were always found greatest in number at the surface layers (0-2 cm) by several workers (Hijink and Kuiper, 1966; Richter, 1969 and Szczygiel and Hasiior, 1972a). Singh and Misra (1968) found Xiphinema and Longidorus spp. quite deep in the soil while Harrison and Winslow (1961), Flegg (1968) found them at upper

surface. Miller (1960, '71), Ferris and Bernard (1961), Potter (1967), O'Ban<sup>n</sup>on et al. (1972) have reported that the following factors influence the vertical distribution of nematodes: root distribution, height of water table, soil moisture, soil temperature, soil texture, rainfall and depth of subsoil. Wallace (1963) related root distribution as the main factor in the vertical distribution of plant-parasitic nematodes. However, Flegg (1968), Singh and Misra (1968) contradicted the observation that root is the main factor in vertical distribution of nematodes. The vertical migration in response to soil water conditions, mechanical composition of soil as well as temperature has also been demonstrated by some of the workers (Lewis and Mai, 1961; Wallace, 1962; Wallace and Greet, 1964 etc.).

The present work was undertaken with a view to study the population fluctuations, seasonal variations, vertical migration, breeding cycle and predator-prey relationship of Hadronchus shakili.

The study was carried out in a plot of about one acre at Company Gardens, Bareilly, Uttar Pradesh on which were plant<sup>n</sup>ed 25 trees of litchi, Litchi chinensis Sonn, in five rows and the soil around these plants harboured the predatory nematode, H. shakili, in abundance. From each row one tree was selected for sampling. Soil samples were collected at three depths, viz.,

0-10, 10-20, 20-30 cm with the help of a hand spade at intervals of 15 days from July, 1977 to June, 1978 (except in late July when it was impossible to collect the samples at middle and lower depths due to water-logging and it has been shown in the figures by dotted lines). The three depths are mentioned in the following text as upper, middle and lower depths. For taking samples an area of about 10 x 10 cm was marked and the total soil of each 10 cm successive depths was taken out and kept separately. This bulk of soil from each depth was mixed up thoroughly and 500 gm of soil was taken out of this bulk into a polythene bag. Thus 15 bags (5 rows of tree x 3 depths) was collected and stored, and was brought to the laboratory.

The five soil samples of the same depth obtained from the 5 rows of trees were composited into one grand sample, were thoroughly mixed and out of this bulk 250 gm soil was taken for processing for the isolation of nematodes. The nematodes were extracted from the soil by modified Baermann funnel method. The water from the bottom of the funnel was taken exactly after 24 hours. This contained nematodes in clear suspension. The nematode suspension obtained as above was transferred to a graduated flask and the nematodes were allowed to settle down at the bottom. The suspension was concentrated to 100 ml by pipetting out the extra volume of water. This 100 ml of nematode suspension was made homogeneous with the help of a bubbler and

10 ml was taken out of this with the help of a pipette and was transferred to a Syracuse counting dish. The nematodes were counted and the counting was repeated thrice and the mean values of the countings noted down. The different juvenile stages and the adults (males and females) were counted separately. Temperature was noted regularly on the day of sampling at each depth. The soil moisture of each sample was determined in the laboratory.

## RESULTS

Soil temperature at three depths was variable at different intervals of this study. The maximum variation ( $10$  to  $26^{\circ}\text{C}$ ) occurred in the upper depth of soil in February and April respectively. The soil temperature between the three layers also varied and a maximum of  $4^{\circ}\text{C}$  difference was noted between two adjacent layers. During winter (November to February), the temperature was higher at the two lower depths, but in summer (March to May) it was just reverse (Fig. XXVII, A). Soil moisture also varied during different seasons and at different depths. The highest (24%) of soil moisture was recorded in July, 1977 and the lowest (6%) in April, 1978 (Fig. XXVII, B).

The studies on population fluctuations of nematodes from July, 1977 to June, 1978 showed that they were abundant during

the monsoon period occurring in maximum numbers in the month August. Afterwards, the population gradually declined with a corresponding fall in the soil moisture. The lowest level of population was reached in April (Fig. XXVIII) where the temperature was maximum and the soil moisture was minimum (Fig. XXVII A & B).

The distribution of worms varied greatly throughout the year especially in the upper and middle depths. The largest number of nematodes were recovered in the upper depth soil while the lowest concentration was found at the lower depth (Fig. XXVIII). The relative distribution of different juvenile stages and the adults at three depths was found to be almost same (Fig. XXIX & XXX). The maximum concentration of H. shakili was noticed at the upper depth from May to October, when the soil moisture and temperature were 17-23% and 16-23°C respectively, except in late July when the entire field was water-logged due to heavy rain (Fig. XXVIII). From October onwards they started migrating downwards and concentrated at middle depth until April, with a gradual decline of 6% in moisture (except in February when the moisture was 20-23% due to rain). Again an upward migration took place from May onwards when the plot was irrigated. The least migration was noticed in the lower depth where there was least variation in temperature as well as moisture (Fig. XXVIII).

The breeding season of Hadronchus shakili started from the month of March. The maximum number of uterine eggs (0.44 eggs

per female) were recorded in early September while the lowest numbers in October (0.04 per female). From late October until late February no eggs were found in the uteri (Fig. XXXI).

The first stage juveniles though present throughout the year increased in number as the breeding season started and reached its peak (46% of the total population of H. shakili) in early September. The lowest level (8%) was recorded in February (Fig. XXXII, A). The highest concentration of the second stage juveniles (27%) was recorded in January, i.e., about five months after the peak period of first stage juveniles, and its lowest count was 12% in May (Fig. XXXII, A). The population of the third stage juveniles varied from 14 to 22% of the total population of H. shakili in June and March respectively (Fig. XXXII, A). The population of the fourth stage juveniles was highest (43%) in February and lowest (7%) in August. On the basis of above observations it can be inferred that the population of fourth stage juvenile was highest when the population of the first stage juveniles was lowest and vice versa (Fig. XXXII). The peak in the population of adults (36% of the total population of H. shakili) occurred about five months later to the peak of fourth stage juveniles, i.e., in June with their lowest count (8%) in October (Fig. XXXII, E). The <sup>male</sup>female ratio was almost constant (1:1) throughout the seasons.

The observation, made on total population of prey (other nematodes) along with the individual populations of Trichodorus sp. Thornenema sp. and Hemicriconemoides sp., to the population of H. shakili showed an inverse relationship (Fig. XXXIII, A & B). The population of Trichodorus sp. increased from August until March as the population H. shakili decreased. In case of Thornenema sp., its population increased as the population of H. shakili decreased and vice versa. The population of Hemicriconemoides sp. (occurring only at middle and lower depths) decreased as the number of H. shakili at the respective depths increased and vice versa (Fig. XXXIII, A). The trend, in the total population of prey was also found almost parallel to the population of Trichodorus sp. (Fig. XXXIII, B).

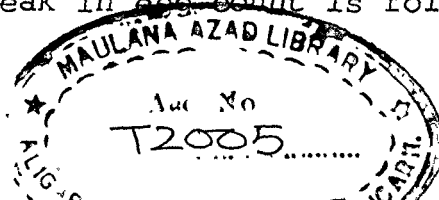
#### DISCUSSION

The results of the present study on Hadronchus shakili support the findings of Kable and Mai (1968), Wallace (1971), O'Bannon et al. (1972) and others that seasonal fluctuations in the nematode population is governed mainly by the variations in the soil moisture. The present results also support the findings of Griffin and Darling (1964) that the minimum population of nematodes is found when maximum soil temperature and minimum soil moisture are encountered.



The <sup>higher</sup> fluctuations in the nematode population at the upper and middle depths and least at the lower depths is correlated with the variations in soil moisture and temperature at the respective depths. This is in accordance with the observations of Griffin and Darling (1964). The sharp decline in the population of H. shakili during late July and its subsequent increase in August at upper depths can be attributed to water-logging. Further, the migration of nematodes from the upper depth to the middle from October to April is due to corresponding decrease in soil moisture in the upper depth. During May after irrigation their migration to the upper depth is again due to soil moisture. This observation supports the findings of Wallace (1962), Wallace and Greet (1964). In spite of optimum moisture (20-23%) at upper depth in the month of February, the nematode density was higher at middle depth. Most probably the upward migration was prevented due to a low temperature ( $10^{\circ}\text{C}$ ) at upper depth in February. This confirms the observations of Lewis and Mai (1961) that temperature affects nematode migration.

The gradual increase in the population of adults and a corresponding decrease in the population of the fourth stage juveniles preceeding the breeding season is due to moulting of the fourth stage juveniles to become adults. The sharp decline in the population of adults in October is perhaps due to the death of spent females. The peak in egg count is followed by an



increase in the population of first stage juveniles. This suggests that the eggs have hatched promptly during this period. Further, the peaks in the population of other stages is not followed by the peak of first stage juveniles which suggests that the span of life of each stage is variable, extending for several months. The continuous presence of uterine eggs from March to October with its peak in September along with the total absence of such eggs from late October until late February clearly indicates that H. shakili breeds only once in a year.

The sharp decline in the total population of prey along with the individual populations of Trichodorus sp. in April is probably due to the dry weather. Their further reduction in May when the soil moisture is sufficient and the number of predator is relatively high appears to be due to over predation. The decline in the population of Hemicriconemoides sp. in April when the predator density was highest may be attributed to the interaction between prey and predator. In case of Thornenema sp. the results are similar to that of Trichodorus sp. except that there is no such sharp decline in the population in April. Thus the population fluctuations of prey is inversely related to the population of predator under normal conditions and it shows that there is a clear antagonism between the prey and the predator (Fig. XXXIII). Further, the results show that the studies on such interactions should be undertaken in relation to the specific

species or genera of nematodes instead of the total population of prey which may not give proper results. This might be due to the diversity in the mode of reproduction of different species, especially in case of free-living nematode, where the life cycle is usually short and the rate of reproduction is tremendous. However, one thing that becomes quite evident from the present study is that even a slight increase in the population level of the predator, H. shakili caused a sharp decline in the population of prey (other nematodes) and vice versa.

## EFFECTS OF CHEMICALS

The toxicity of different chemicals to the various nematode species including the juvenile stages was demonstrated by Fenwick (1957); Goring (1957); Moje (1959); Moje and Thomason (1963); Bird and McGuire (1966); Marks et al. (1968); Esser (1972); Husain and Masood (1974); Keeth (1974), etc. Stephenson (1945) studied the effects of fatty acids on Rhabditis terrestris Stephenson, 1942 and found that formic acid was relatively more toxic of all the acids tested. Johnston(1959) and Banage and Visser (1965) also studied the effects of these acids on Tylenchorhynchus martini Fielding, 1956 and Dorylaimus sp. respectively and found out just the reverse of the above that formic acid was less toxic to these species in comparison with other acids. Johnston (1959) concluded that the effectiveness of the acids increases with their molecular weight. Banage and Visser (1965) found that undissociated acid molecule is the chief toxic factor.

The hydrogen ion concentration (pH) is an important abiotic factor influencing the nematodes directly or indirectly (Robinson and Neal, 1956; Loewenberg et al., 1960; Johnson and Viglierchio, 1961; etc.). Soroczan (1969) while working on the survival rate of larvae and the adults of Rhabditis sp. under different pH gradients and at different concentrations of mineral salts concluded that pH 7.5 to 8.8 and lower salt

concentrations were more suitable for the nematodes. Pitcher and McNamara (1972) working on Pratylenchus penetrans (Cobb, 1977), Xiphinema diversicaudatum (Micoletzky, 1927) and Aphelenchoides ritzemabosi (Schwartz, 1911) found out that the first species was susceptible to silver ions, second to cupric ions, while the third one was neither effected by silver nor cupric ions. Jairajpuri et al. (1974) also studied the effects of pH and mineral salt concentrations on the survival of Hoplolaimus indicus Sher, 1963; Helicotylenchus indicus Siddiqi, 1963; Xiphinema basiri Siddiqi, 1959 and Mylonchulus minor (Cobb, 1893). They concluded that copper sulphate was most toxic of all the salts that were tested. Xiphinema basiri and Mylonchulus minor were found to be more susceptible to slight changes in their optimal pH while the other two species showed a greater degree of tolerance.

In the present work the effects of pH, fatty acids (formic, butyric, propionic and acetic) and mineral salts ( $\text{CuSO}_4$ , KCl,  $\text{KNO}_3$  and  $\text{K}_2\text{CO}_3$ ) on the survival of the adults and juveniles of Hadronchus shakili has been investigated. The changes that occur in the movement and in their postures during exposure to chemicals were also noted.

The nematodes were rinsed repeatedly in distilled water soon upon extraction. The adults and the different juvenile

stages were kept separately in petri-dishes. The solutions (acids, salts and phosphate buffers) were also prepared in the distilled water.

To study the effects of acids, the adults and the juvenile stages were exposed to eight concentrations of acids (1N, 0.8N, 0.6N, 0.4N, 0.2N, 0.1N, 0.01N and 0.001N). Similarly, for each salt five molar concentrations were taken (0.4M, 0.3M, 0.2M, 0.1M and 0.05M). For determining the effects of hydrogen ion concentration the worms were treated with the phosphate buffers of pH: 2.2, 3.0, 4.0, 5.0, 5.4, 5.6, 5.8, 6.0, 6.4, 7.0, 7.4 and 8.0. After recording the immobilization time of the nematodes at a particular pH, the immobilized worms were transferred to cavity-blocks containing tap-water in which their survival time was noted. The survival time in tap-water and buffer solutions was noted in order to determine the degree of susceptibility of the two sexes and the various juvenile stages separately. Based on their effects on the nematodes the pH was graded as follows: highly toxic, less toxic and optimum.

After treatment with the chemicals when the nematodes became inactive even after repeated proddings with a needle, they were presumed immobilized or dead. The immobilization time was recorded with the help of a stop-watch. The observations were taken at intervals of five seconds in the first hour, after

10 minutes in the second hour, and afterwards at intervals of one hour. If the nematodes became dead between any two readings, a mean of the two was considered as the immobilization time. Those nematodes which survived longer than 24 hours in any solution were transferred to fresh media of the same solution. Each treatment was replicated ten times.

## RESULTS

### EFFECT OF pH (TABLE VI, FIG. XXXIV, A & B)

The survival of adults and juveniles was found to be effected by pH. pH 2.2 to 4.0 proved highly toxic and the survival at this range in the phosphate buffer was 2.5 to 7.1 min for females and 1.8 to 6.1 min for males. The first stage juveniles showed minimum survival time (0.6 to 2.7 min) while the fourth stage showed maximum (1.5 to 6.1 min). In case of second and third stage juveniles the survival time was nearly same, i.e., 1.1 to 4.0 min and 1.0 to 4.0 min respectively. Upon becoming immobilized in the buffer solution, the worms were transferred to tap-water, but they showed no revival except at pH 4.0 in which the adults and the fourth stage juveniles revived and lived for 12 to 12.7 hr and the second and third stages for about 5.1 hr only. The span of survival for the two sexes and the juvenile stages was almost same at pH 5.0 to 5.6, but after

transfer to tap their survival time was different at each pH being 40.43 hr for females, 36 to 41 hr for males and 36 to 40 hr for the fourth stage juveniles. For the third, second, and first stages it varied from 5.5 to 11.5 hr, 5 to 7.8 hr and 2 to 7.1 hr respectively. At pH 5.8 the survival period for adults as well as juveniles in buffer solutions and tap-water was maximum (10 min, 9.5 min, 8.8 min, 6.0 min, 5.6 min and 4.1 min in buffer solutions and 61 hr, 58 hr, 52 hr, 13.5 hr, 13.3 hr and 9.3 hr in tap-water for females, males, fourth, third, second and first stage juveniles respectively). The survival time for these nematodes in buffer solutions was almost same at pH 5.8 and 6.0 but it differed in tap-water. At pH 6.4 or more, the survival time of these stages decreased both in the buffer solutions and tap-water, the minimum being at pH 8.0. In tap-water at pH range 6.4 to 8.0, the survival time was 19.2 to 29 hr for females, 16.3 to 26 hr for males, 16 to 24 hr for the fourth stage, 9 to 11.5 hr for the third stage, 6 to 9.8 hr for second stage and 5.3 to 5.5 hr for the first stage juveniles.

From the above observations it is evident that all the stages have almost same optimum pH but their survival time is different. The females show maximum tolerance to pH while the first stage juveniles show minimum. On the basis of the degree of tolerance the following series for these stages could be established: females > males > fourth stage > third stage > second



stage, first stage juveniles. These may further be grouped into three: i) adults and fourth stage juveniles, ii) second and third stage juveniles, and iii) first stage juveniles. The lower survival time for adults and juveniles at pH 7.4 to 8.0 shows that this species can not survive even at low alkaline medium. Moreover, the maximum span of survival for adults and juveniles at pH range 5.8 to 6.0 shows that this is the optimum pH range for this species. Based on the survival time of nematodes, the pH range (2.2 to 8.0) may be divided in three groups: i) Highly toxic (2.2 to 4.0 and 7.4 to 8.0), ii) Less toxic (5.0 to 5.6 and 6.4 to 7.0), iii) optimum (5.8 to 6.0).

#### EFFECT OF MINERAL SALTS (TABLE VII, FIG.XXXV)

##### Potassium carbonate (Fig. XXXV, A):

This salt was least toxic of all the salts. The survival time at concentrations 0.4 to 0.2M was 10 to 40 min for females, 7 to 36 min for males and the fourth stage juveniles, and 5 to 29 min for the third stage. In case of the second and first stage juveniles the survival period was 3 to 20 min and 3 to 12 min respectively. At lower concentrations 0.1 to 0.05M, the maximum survival time was recorded 1.7 to 2.5 hr for adults, 1.5 to 2.2 hr for fourth stage, 0.8 to 1.9 hr for third stage, and about 0.5 to 1 hr for the second and first stage juveniles.

Potassium chloride (Fig. XXXV, B):

The survival time for adults and fourth stage juveniles at concentrations 0.4 to 0.1M was 2 min to 1 hr. For others the survival at these concentrations was 2 to 42 min for the third stage; 1 to 38 min for second stage and 1 to 27 min for the first stage juveniles. At the lowest concentration of 0.05M, the first stage survived up to 42 min while the second and third stages for 1.2 and 1.4 hr respectively. In case of adults and fourth stage juveniles the survival time was about 2 hr.

Potassium nitrate (Fig. XXXV, C):

This salt was found to be more toxic than potassium chloride. The maximum survival time for the adults and fourth stage juveniles was 37 min and 27 min, while 16<sup>min</sup> and 19 min for the second and third stages and only 9 min for the first stage juveniles. At highest concentration of 0.4M, the survival time was 3 min for adults and fourth stage, 2 min for the third stage and about 1 min for the second and first stage juveniles.

Copper sulphate (Fig. XXXV, D):

Of all the salts, this was most toxic for the nematodes. At 0.4 to 0.2M concentrations, the survival time was 2 to 4 min for females, 2 to 3 min for males and fourth stage, 1 to 2 min for third and second stages and 0.5 to 1 min for the first stage

juveniles. At lower concentrations of 0.1 to 0.05M the survival time was 11 to 15 min for females, 10 to 15 min for males and 7 to 13 min for the fourth stage juveniles. In case of third and second stage juveniles it was 6 to 9 min and 4 to 8 min respectively. The minimum survival time of 3 to 5 min was noted for the first stage juveniles.

The adults and juveniles of H. shakili when exposed to various concentrations of salts solutions showed a varying degree of susceptibility. The survival time of each juvenile stage and the two sexes was found to be different at same concentration. The first stage juveniles showed minimum survival at every concentration while it was maximum for the females. The degree of toxicity of these salts is in the following order: Copper sulphate > potassium nitrate > potassium chloride > potassium carbonate.

#### EFFECT OF FATTY ACIDS (TABLE, VIII, FIG. XXXVI and XXXVII)

##### Acetic acid (Fig. XXXVI, A):

This acid was found to be least toxic of all the acids tested. At concentrations of 1.0 to 0.01N, the survival time was 4 to 51 min for females, 3 to 47 min for males and fourth stage, 2 to 39 min for third stage, 2 to 32 min for second stage and 1 to 19 min for the first stage juveniles. At the lowest concentra-

tion of 0.001N, the survival of each stage was longer: 28.3 hr, 27.2 hr, 26.3 hr, 21.6 hr, 21 hr and 13 hr for the females, males, fourth, third, second and the first stage juveniles respectively.

Propionic acid (Fig. XXXVI, B):

The survival time at higher concentrations (1.0 to 0.1N) was 3 to 15 min for adults and the fourth stage, 2 to 10 min for third and second stage juveniles. In case of first stage juveniles the survival was 1 to 3 min only. At concentrations of 0.01 to 0.001N, the females survived for 0.7 to 26.7 hr. Though the survival at concentration 0.01N for males, fourth and third stage juveniles was almost same that is about 0.5 hr, but at lowest concentration (0.001N) it differed. The third stage juveniles survived for 19.8 hr while male and the fourth stage juveniles for 26.5 hr and 26 hr respectively. At concentrations (0.01 to 0.001N) the survival for second and first stage juveniles was 0.3 to 19.4 hr and 0.2 to 12.7 hr respectively.

Butyric acid (Fig. XXXVII, A):

In this acid the survival time of the nematodes was almost same as in propionic acid except that the first stage juveniles showed comparatively greater degree of susceptibility. The survival time at various concentrations for this stage was <1 min

at 1.0 to 0.4N, 1 to 11 min at 0.2 to 0.01N and about 10.5 hr at 0.001N.

Formic acid (Fig. XXXVII, B):

This acid was highly toxic at higher concentrations. At lower concentration of 0.001N the survival time for each stage was longer as in case of butyric, propionic and acetic acids. The females showed a comparatively lesser degree of susceptibility. The survival of females at concentrations 1.0 to 0.01N and 0.001N was 1 to 9 min and 22.9 hr while for males and fourth stage juveniles it was  $\angle$  1 to 8 min and 21.7 to 22.1 hr respectively. The survival period at the lowest concentration of 0.001N for the third and second stages was maximum being 18 hr and 17.1 hr respectively. At concentrations 1.0 to 0.01N these stages survived for  $\angle$  1 min to 7 min. The first stage juveniles survived for lesser periods at all concentrations in comparison to other stages. The survival being  $\angle$  1 min to 4 min and 8.7 hr at concentrations of 1.0 to 0.01N and 0.001N respectively.

It is evident from the above observations that the degree of tolerance of various stages to each acid was on the same pattern as those for mineral salts, i.e., the first stage juveniles being most susceptible and the adult females the least. Of all the acids tested, the formic acid proved most toxic. On the basis of their toxicity the following order could be established.

acetic acid / propionic acid / butyric acid / formic acid.

#### GENERAL OBSERVATIONS

The exposure of adults and all the juvenile stages of Hadronchus shakili to various chemicals showed different results. At higher concentrations of acids and at pH (2.2 - 4.0) the worms became highly inactive and showed only a few uncontrolled jerky movements before becoming completely immobilized. At lower concentrations, the worms gradually showed increased activity and apparently looked more agitated. These movements resulted in the formation of spiral postures which were either sinistral or dextral or irregularly spiral. Before the final immobilization or death the nematodes showed some violent movements followed by straightening of the body. Afterwards they ceased to respond to proddings. The males after becoming inactive (or upon death) showed a strongly curved hinder part of their bodies and their spicules were generally protruding out. In females, the hinder part was only slightly curved. Similar observations were recorded in salt solutions also. During the entire period of immobilization the worms showed various postural changes, e.g., spiral shaped, s-shaped, c-shaped, open c-shaped, closed c-shaped. Sometimes postures like figure of '3' or '8' were also noted. In juveniles, no obvious differences either in movement or in posture were observed.

Besides the above, osmotic effects was also evident on the worms. Due to a gradual process of ex~~o~~smosis the body shortened resulting in the shrinkage of the outer cuticle and internal body organs, like intestine, oesophagus etc. During this process, darkening of the tissues also took place. When transferred to water, the worms remained inactive for sometime until they assumed original shape and movement as before the exposure except at 0.001N of acids and pH 2.2 to 3.0. At pH 4.0, only the adults, the fourth and the third stage juveniles resumed activity but the others died.

These observations on motility of nematodes and osmotic effects are similar to those observed by Banage and Visser(1965), Evans and Thomason (1971) and Jairajpuri et al. (1974).

#### DISCUSSION

The present study reveals that H. shakili has a very narrow range of optimum pH like that of Mylonchulus minor (cf. Jairajpuri et al., 1974). The low alkaline medium also proved to be highly toxic. These observations further support the hypothesis of Jairajpuri et al. that sensitiveness of the species to pH seems to be a limiting factor for its distribution and propagation. Among all the salts tested, copper sulphate was most toxic while the potassium carbonate was least. However,

the toxicity of copper sulphate was not as high as was in M. minor.

Of the various fatty acids tested formic acid proved to be most toxic. This observation is similar to that of the Stephenson (1945) for Rhabditis terrestris. The toxicity of butyric, propionic and acetic acids was similar to that given by earlier workers (Stephenson, 1945; Johnston, 1959 and Banage and Visser, 1965). The toxicity of acids at same concentration for different stages and the two sexes also differed and these results agree with the findings of Stephenson (1945) and Johnston (1959).

The susceptibility of various developmental stages to pH, salts and acids show that the juveniles were more susceptible to these chemicals than their adults. The results are similar to the observations of Evans and Thomason (1971) for Aphelenchus avenae. As reported by Esser (1972) for Radopholus similis, the females showed comparatively greater degree of tolerance than their males. Among the juveniles, the first stage was most susceptible while the fourth stage was least.

From the present study it becomes quite evident that these worms are highly sensitive to chemicals and it is almost definite that the chemicals used for killing the plant-parasitic nematodes also kill these useful animals. The destruction of



predatory nematodes is most likely to disturb the natural ecosystem which may lead to further increase in the population of plant-parasitic nematodes instead of bringing it down.

## SUMMARY

The predatory nematode, Hadronchus shakili Jairajpuri, 1969 is a large sized mononch widely distributed in this country especially in the Northern and North-eastern regions. The present work on this nematode species includes study of its gross morphology and histological anatomy and the description of its various juvenile stages. The variability of important taxonomic characters in the adults and the juveniles has been statistically analysed. Comparative development of various organs in juveniles and adults has also been studied in detail followed by observations on the population dynamics of the species. Lastly, the effects of some toxic chemicals were determined.

The morphological studies revealed that the shape of body, lip region and buccal cavity show little variations but the total body length, number of denticles on the subventral walls, vulval papillae, number of ventromedian supplements, tail length etc. are more variable. The size of the buccal cavity is to some extent correlated with the sex and length of the body of the animal.

The anatomical observations were carried out with particular reference to musculature, digestive and reproductive organs. The cross sections at various levels of the body show

four lateral chords and four platymyarian somatic muscle cells per quadrant. Well developed labial muscles are attached to <sup>the</sup> lips and stomatal muscles to the stoma. The vulval muscles are also of two types, the constrictor vulvae and the dilator vulvae. The oviduct-uterus junction is provided with prominent sphincter muscles. The copulatory muscles, accessory copulatory muscles, spicular muscles, and caudal copulatory muscles are also well developed. In females, the anal region has 2-3 anal muscle bands while in males there is a single band. The digestive organs include hexaradiate vestibulum, tri-radial buccal cavity, muscular oesophagus and tuberculate type of oesophago-intestinal junction. The lumen of oesophagus is tri-radiate and heavily sclerotized, the thickenings serving as points of attachment for the radial muscle fibers. The intestine has 6-8 cells in circumference. The females are amphidelphic and each sexual branch consists of a reflexed ovary, an oviduct with narrow distal and an enlarged sac-like proximal part and a long flexible uterus. The males are diorchic, each testis is followed by a vas deferens, and an ejaculatory duct joining the cloaca. The copulatory structures include paired spicules, a gubernaculum and lateral accessory pieces.

The important characters for separating the different juvenile stages are, total body length; length and width of

buccal cavity; number, orientation and position of teeth in the buccal cavity; length of oesophagus; size of the genital primordia etc. The first stage juveniles are always devoid of denticles on the vertical subventral walls while in the remaining stages these are present. The juveniles can easily be distinguished from their adults by the presence of a spare tooth at the base of the functional dorsal tooth.

The analysis of variability of some important morphological characters of the adults and juveniles revealed that almost all the characters are variable to some extent. The length and width of buccal cavity are, however, least variable. The position of nerve ring, anal body-width and length of spicules are also not much variable. The highly variable characters include the position of amphid apertures, length of female sexual branches, tail, rectum etc. The vulva position is positively correlated with the total body length while the lengths of genital branches are negatively correlated. Of all the allometric characters, the value of  $V$  is least variable and  $G_1$  and  $G_2$  exhibit maximum variability. The length of buccal cavity, oesophagus and tail are independent of body length. The characters in juveniles vary almost to the same extent as in the adults.

The buccal cavity is formed anew during the process of moulting. A total of four moults occur. In the late phase of

each moult there are always three dorsal teeth one of these is cast off along with the old buccal cavity, the second one becomes the functional tooth of the stage to follow, while the third one is the spare tooth of the future stage. The development of the buccal cavity is more pronounced in the adults than in the juveniles. The development of oesophagus and tail in relation to total body length is more in juveniles than the adults. The genital primordia in the first stage juveniles consists of two oval bodies connected by a cellular strand. Each oval body is with one germinal nucleus and two somatic nuclei. The cap cell nuclei are derived from the division of the somatic nuclei at the distal and proximal ends of the primordia and do not divide again. The germinal nuclei divide for the first time in the third moult while the somatic nuclei in the second moult. The somatic nuclei proliferate in the middle region of the primordia to form the gonoduct. The epidermal nuclei are derived from the somatic nuclei at the opposite ends of the primordia and form the epidermal walls of the testes and ovaries. The vagina is formed by the specialized ventral chord nuclei, whereas the development of spicules and gubernaculum takes place from the spicular primordia in the anal region <sup>which is</sup> distinguishable in the third stage male juveniles. The sex differentiation occurs in the second stage juveniles when specialized ventral chord nuclei make their appearance in the female juveniles. The development

of the genital tract and ovary in various seasons also differed. The frequency of egg production by the anterior ovary was much higher than that of the posterior ovary (1:2).

Observations were made on the fluctuations in the population of this nematode in a plot of about one acre at the Company Gardens, Bareilly, Uttar Pradesh from July 1977 to June 1978. For this purpose, soil samples at depths of 0-10, 10-20 and 20-30 cm were collected twice a month. The nematodes were extracted separately from these three types of samples and were counted and classified age-wise. The seasonal fluctuations in the population was found to be chiefly governed by the moisture contents of the soil, because the maximum level of the population was noted during the peak monsoon period (August) while the minimum during the dry period (April). The data on the vertical migration revealed that this species occurs abundantly in the surface layer from May to October (Except for a brief period during July when the entire field was water-logged). From October onwards they start to migrate downwards and concentrate at 10-20 cm depth until April. At the depth of 20-30 cm the population was always found to be low. On the basis of abundance of juveniles and the gravid females, the breeding period of this nematode appears to begin in March and extends up to October, but apparently they bred only once a year. An inverse relationship exists between the population of

this mononch and those of its prey, the soil-inhabiting nematodes including plant parasites. This suggests a clear antagonism between the prey and predator.

The observations on the effects of pH, mineral salts and fatty acids showed that these chemicals are toxic to this species. The high susceptibility to the pH gradients ranging 2.2 to 4.0 and 6.4 to 8.0 showed that these nematodes can not survive either in high acidic media or low alkaline media. The maximum life span at pH 5.8 to 6.0 shows that this is the optimum pH range. The mineral salts of concentrations ranging from 0.4M to 0.05 M showed different effects at different concentrations. The rate of survival was lower at higher concentrations. Copper sulphate was most toxic, while

potassium chloride was least. The fatty acids ranging from 1N to 0.001N also proved highly toxic, but the toxicity at lowest concentration of 0.001N was less. Formic acid proved highly toxic. The adults in general were more tolerant than the juveniles. The adult females were least susceptible while the first stage juveniles were most. The exposure to various chemicals also brought about changes in the posture and movement of the nematodes.

## REFERENCES

- ABOUL-EID, H. Z. (1969). Histological anatomy of the excretory and reproductive systems of the Longidorus macrosoma. Nematologica, 15: 437-450.
- AHMAD, I. and JAIRAJPURI, M. S. (1979). Developmental biology of Chiloplacus symmetricus. Indian J. Nematol., 7: 123-139.
- ANDERSON, R. V. and DARLING, H. M. (1964). Embryology and reproduction of Ditylenchus destructor Thorne, with emphasis on gonad development. Proc. helminth. Soc. Wash., 31: 240-256.
- AZMI, M. I. and JAIRAJPURI, M. S. (1978). Morphometric and allometric variations in the adults and juveniles of Helicotylenchus indicus. Indian J. Nematol., 6: 13-22.
- BAJAJ, H. K. and JAIRAJPURI, M. S. (1977). Variability within Xiphinema insigne populations from India. Nematologica, 13: 617-632.
- BAJAJ, H. K. and JAIRAJPURI, M. S. (1977). Statistical analysis of variability in a population of Xiphinema basiri Siddiqi, 1959. Nematol. Medit., 5: 269-280.
- BANAGE, W. B. and VISSER, S. A. (1965). The effect of some fatty acids and pH on a soil nematode. Nematologica, 11: 255-262.



- BAQRI SAMER, Z. and JAIRAJPURI, M. S. (1974). Studies on Mononchida of India V. Some observations on the morphology of Mononchus aquaticus Coetzee, 1968 with remarks on its status. Indian J. Nematol., 2: 105-116.
- BINGEFORS, S. (1957). Studies on breeding red clover for resistance to stem nematodes. Växteodling, 8: 1-123.
- BIRD, G. W. (1966). Influence of host and geographical origin on population of Trichodorus christiei, Nematologica, 12: 88 (Abstr.).
- BIRD, G. W. and MAI, W. F. (1965). Plant species in relation to morphometric variations of the New York population of Trichodorus christiei, Nematologica, 11: 34 (Abstr.).
- BIRD, G. W. and MAI, W. F. (1967). Morphometric and allometric variations of Trichodorus christiei. Nematologica, 13: 617-632.
- BIRD, A. F. and McGUIRE, R. J. (1966). The effect of anti-metabolites on the growth of Meloidogyne javanica. Nematologica, 12: 637-640.
- BIRD, K. (1973). Nematodes of Lake Belaton IV. Seasonal qualitative and quantitative changes. Annal. Biol. Tihany., 40: 135-158.
- BRZESKI, M. (1963). Morphological studies on Eudorylaimus silvaticus Brzeski (Nematoda, Dorylaimidae). Bull. Acad.

- Pol. Sci. Cl. II. S'er. Sci. biol., 11: 133-136.
- CALPHAM, P. A. (1930). On variations in size of the nematodes worm Rhabditis succaris n. sp. produced by different culture media. J. Helminth., 8, 211-222.
- CASSIDY, G. (1931). Some Mononchs of Hawaii. Hawii. Plrs. Rec., 35: 305-339.
- CHIN, D. A. (1977). Embryonic and post-embryonic development of Cylindrocorpus longistoma (Nematoda : Cylindrocorporidae). Nematologica, 23: 62-70.
- CHITWOOD, B. G. and CHITWOOD, M. B. (1950). An introduction to Nematology. Section I. Anatomy. Chapter X, pp. 136-159, Chapter VIII, pp. 116-125 Baltimore Monumental Printing Company.
- CHUANG, S. H. (1962). The embryonic and post-embryonic development of Rhabditis teres (A. Schneider). Nematologica, 7: 317-330.
- CLARK, W. C. (1960a). The oesophago-intestinal junction in the mononchidae (Enoplida, Nematoda). Nematologica, 5: 178-183.
- CLARK, W. C. (1960b). Redescription of Mononchus truncatus Bastian, M. papillatus Bastian, and Prionchulus muscorum (Dujardin) (Enoplida, Nematoda). Nematologica, 5: 184-198.

- CLARK, W. C. (1960c). New Zealand Mononchidae (Enoplida, Nematoda) I. The genera Mononchus Bastian and Prionchulus Cobb. Nematologica, 5: 260-274.
- CLARK, W. C. (1960d). The Mononchidae (Enoplida, Nematoda) of New Zealand II. The genus Iotonchus (Cobb, 1916) Altherr, 1950. Nematologica, 5 : 260-274.
- CLARK, W. C. (1960e). The Mononchidae (Enoplida, Nematoda) of New Zealand III. A review of the genus Cobbonchus Andrassy, 1958 with descriptions of new species. Nematologica, 5 : 275-285.
- CLARK, W. C. (1961a). The Mononchidae (Enoplida, Nematoda) of New Zealand IV. The genus Mylonchulus (Cobb, 1916) Pennak, 1953. Nematologica, 6 : 1-6.
- CLARK, W. C. (1961b). A revised classification of the order Enoplida (Nematoda). N. Z. Jl Sci., 4 : 123-150.
- CLARK, W. C. (1962). Evolution within the family Mononchidae (Enoplida, Nematoda). Nematologica, 7 : 251-255.
- CLARK, W. C. (1963). Notes on the Mononchidae (Nematoda) of the New Zealand region with the descriptions of new species. N. Z. Jl Sci., 6 : 612-632.
- COBB, N. A. (1916). Notes on new genera and species of nematodes. 4. Subdivisions of Mononchus. J. Parasit., 2 : 195-196.

- COBB, N. A. (1917). The Mononchs (Mononchus Bastian, 1866).  
A genus of free-living predatory nematodes. Soil Sci.,  
3: 431-486.
- COBB, N. A. (1920). Transfer of Nematodes (Mononchs) from  
place to place for economic purposes. Sci. n. S.,  
51: 640-641.
- COHN, E. and MORDECHAI, M. (1974). Experiments in supressing  
citrus populations by use of a marigold and a  
predaceous nematode. Nematol. Medit., 2 : 43-53.
- COOMANS, A. (1962). Morphological observations on Rotylenchus  
goodeyi Loof and Oostenbrink, 1958. I. Redescription  
and variability. Nematologica, 7 : 203-215.
- COOMANS, A. (1964). Structure of female gonads in member of  
Dorylaimina. Nematologica, 10 : 601-622.
- COOMANS, A. and LIMA, M. B. (1965). Description of Anatonchus  
amiciae n. sp. (Nematoda : Mononchidae) with observations  
on its juvenile stages and anatomy. Nematologica, 11 :  
413-431.
- COOMANS, A. and LOOF, P. A. A. (1970). Morphology and taxonomy  
Bathyodontina (Dorylaimida). Nematologica, 16 : 180-196.
- DASGUPTA, D. R., NAND, S. and SESHADRI, A. R. (1970). Culturing,  
embryology and life history studies on the lance nematode  
Hoplolaimus indicus. Nematologica, 16 : 235-248.

- DE MAN, J. G. (1884). Die freiinder reinen Erade und im Süssan Wasser Lebenden Nematoden niederlandischen Fauna. Eine systematische-faunistische Monographie, Leiden. 206 pp.
- ESSER, R. P. (1963). Nematode interactions in plates of non-sterile water agar. Proc. Soil Crop Sci. Soc. Fla., 23 : 121-138.
- ESSER, R. P. (1972). Effect of Sodium hypochlorite concentrations on selected genera of nematodes. Proc. helminth. Soc. Wash., 39 : 108-114.
- EVANS, A. A. F. and THOMASON, I. J. (1971). Ethylene dibromide toxicity to adults larvae and moulting stages of Aphelenchus avenae. Nematologica, 17 : 243-254.
- FENWICK, D. W. (1957). Preliminary studies on the effect of ethylene dibromide fumigation on the hatachability of Heterodera rostochiensis (Woll.). Nematologica, 2 : 241-249.
- FERRIS, V. R. and BERNARD, R. L. (1961). Seasonal variations of Nematode populations in soyabean field soil. Plant Dis. Repr., 45 : 789-793.
- FISHER, J. M. (1965). Studies on Pratylenchus nanus. I. Effects of variation in environment on several morphometric charactors of adults. Nematologia, 11 : 269-279.

- FLEGG, J. J. M. (1967). Genital tract development in Xiphinema vuittenezi. Nematologica, 13 : 640-641.
- FLEGG, J. J. M. (1968). The occurrence and depth distribution of Xiphinema and Longidorus species in south eastern England. Nematologica, 14 : 189-196.
- GERAERT, E. (1968). Morphometric relations in nematodes. Nematologica, 14 : 171-183.
- GERAERT, E. (1978a). On growth and form in Nematoda : I. Oesophagus and body-width in Tylenchida. Nematologica, 24 : 137-158.
- GERAERT, E. (1978b). On growth and form in Nematoda : II. Oesophagus and body-width in Dorylaimida. Nematologica, 24 : 347-360.
- GERAERT, E. (1979). Growth and form in Nematodes : III. Comparison of oesophagus and body shape. Nematologica, 25 : 1-21.
- GOODEY, J. B. (1952). The influence of host on dimensions of plant-parasitic nematode Ditylenchus destructor. Ann. app. Biol., 39 : 468-474.
- GORING, C. A. I. (1957). Factors influencing diffusion and nematode control by soil fumigants. A.C.D. Information Bulletin, 110. Dow Chemical Co., Midland, Michigan.

- GRIFFIN, G. D. and DARLING, H. M. (1964). An ecological study of Xiphinema americanum Cobb in an ornamental spruce nursery. Nematologica, 10 : 471-479.
- GROOTAERT, P. and MAERTENS, D. (1976). Cultivation and life cycle of Mononchus aquaticus. Nematologica, 22 : 173-181.
- GROOTAERT, P. and Wyss, U. (1979). Ultrastructure and function of the anterior feeding apparatus in Mononchus aquaticus. Nematologica, 25 : 163-173.
- GUNDY, S. D. VAN. (1958). The life history of the citrus nematode, Tylenchulus semipenetrans Cobb. Nematologica, 3 : 284-294.
- HARRISON, B. D. and WINSLOW, R. D. (1961). Laboratory and field studies on the relation of arbis mosaic virus to its nematode vector Xiphinema diversicaudatum (Micoletzky). Ann. app. Biol., 49 : 621-633.
- HIJINK, M. J. and KUIPER, K. (1966). Waarnemingen over de Verdeling van aaltijes in de grond (Observations on the distribution of nematodes in soil). Meded. Rijksfac. Landbouwwetenschappen Gent, 31 : 558-571.
- HIRSCHMANN, H. (1962). The life cycle of Ditylenchus triformis with emphasis on post-embryonic development. Proc. helminth. Soc. Wash., 29 : 30-43.

- HIRSCHMANN, H. and TRIANTAPHYLLOU, A. C. (1967). Mode of reproduction and development of reproductive system of Helicotylenchus dihystra. Nematologica, 13 : 558-574.
- HOLLIS, J. P. and FIELDING, M. J. (1955). Studies on population dynamics of plant nematodes. Ann. Rep. La. Agric. exp. stn., 1954-1955, 16 pp.
- HUSCAIN, S. I. and MASOOD, A. (1974). Effect of certain organic chemicals on the mortality of Helicotylenchus sp. Indian J. Nematol., 3 : 134-137.
- JAIRAJPURI, M. S. (1969). Studies on Mononchida of India I. The genera Hadronchus, Iotonchus and Miconchus and a revised classification of Mononchida, New Order. Nematologica, 15 : 557-581.
- JAIRAJPURI, M. S. (1970a). Studies on Mononchida of India.II. The genera Mononchus, Clarkus n. gen. and Prionchulus (Family Mononchidae Chitwood, 1937). Nematologica, 16 : 213-221.
- JAIRAJPURI, M. S. (1970b). Studies on Mononchida of India.III. The genus Mylonchulus (Family Mylonchulidae Jairajpuri, 1969). Nematologica, 16 : 434-456.
- JAIRAJPURI, M. S. (1971). Studies on Mononchida of India.IV. The genera Sporonchulus, Bathyodontus and Oionchus (Nematoda). Nematologica, 17 : 407-412.



- JAIRAJPURI, M. S., AZMI, M. I. and BAJAJ, H. K. (1974).  
Effect of pH and salt concentrations on the survival  
of Hoplolaimus indicus, Helicotylenchus indicus,  
Xiphinema basiri and Mylonchulus minor. Indian J.  
Nematol., 4 : 171-181.
- JAIRAJPURI, M. S. and AZMI, M. I. (1978). Some studies on  
the predatory behaviour of Mylonchulus dentatus.  
Nematol. Medit., 6 : 205-212.
- JAIRAJPURI, M. S. and BAJAJ, H. K. (1978). Observations on  
the biology of Xiphinema basiri and X. insigne.  
Rev. Nematol., 1 : 227-239.
- JOHNSON, R. N. and VIGLIERCHIO, D. R. (1961). The accumula-  
tion of plant parasitic nematode larvae around  
carbondioxide and oxygen. Proc. helminth. Soc. Wash.  
28 : 171-174.
- JOHNSTON, T. M. (1959). Effect of fatty Acid mixtures on  
the rice stylet Nematode (Tylenchorhynchus martini  
Fielding, 1956). Nature, Lond., 183 : 1392.
- KABLE, P. F. and MAI, W. F. (1968). Influence of soil  
moisture on Pratylenchus penetrans. Nematologica,  
14 : 101-122
- KELTH, D. P. (1974). The effect of nematicides on the feeding  
posture and dispersal of Aphelenchus avenae.  
Nematologica, 20 : 107-118.

- KHAN, A. M., ADHMI, A. and SAXENA, S. K. (1971). Population changes of some stylet bearing nematodes associated with mango (Mangifera indica). Indian J. Nematol., 1 : 99-105.
- LEWIS, G. D. and MAI, W. F. (1961). Over wintering and migration of Ditylenchus dipsaci in organic soil southern New York. Phytopathology, 50 : 341-343.
- LOEWENBERG, J. R., SULLIVAN, T. and SCHUSTER, M. L. (1960). The effect of pH and minerals on the hatching and survival of Meloidogyne incognita larvae. Pytopathology, 50 : 215-217.
- LOOF, P. A. A. and MAAS, P. W. TH. (1972). The genus Xiphinema (Dorylaimida) in Surinam. Nematologica, 18 : 92-119.
- MARKS, C. F., THOMASON, I. J. and CASTRO, C. E. (1968). Dynamics of the permeation of nematodes by water, nematocides and other substances. Exp. Parasit., 22 : 231-337.
- MAUPAS, E. (1899). La mue et l'enkystement chez les nematodes. Arch. Zool. Exper. et Gen. 3s. 7 : 563-628.
- MILLER, L. I. (1960). The influence of soil components on the survival and development of sting nematode, Belonolaimus longicaudatus. Rau. Va. J. Sci., 11 : 160 (Abstr.).

- MILLER, L. I. (1971). The influence of soil texture on the survival of Belonolaimus longicaudatus. Phytopathology, 62 : 670-671 (Abstr).
- MOJE, W. (1959). Structure and nematicidal activity of allylic and acetylenic halides. J. Agr. Food Chem., 7 : 702-707.
- MOJE, W. and THOMASON, I. J. (1963). Toxicity of mixtures of 1,2-dibromo-3-chloropropane and 1,2-dibromoethane to the root-knot nematode, Meloidogyne javanica. Phytopathology, 53 : 428-431.
- MULVEY, R. H. (1961a). The Mononchidae: A family of predaceous nematodes I. Genus Myelonchulus (Enoplida : Mononchidae). Can. J. Zool., 39 : 665-696.
- MULVEY, R. H. (1961b). The Mononchidae: A family of predaceous nematodes II. Genus Anatonchus (Enoplida : Mononchidae). Can. J. Zool., 39 : 807-826.
- MULVEY, R. H. (1962). The Mononchidae: A family of predaceous nematodes III. Genus Miconchus (Enoplida : Mononchidae). Can. J. Zool., 40 : 65-81.
- MULVEY, R. H. (1963a). The Mononchidae: A family of predaceous nematodes IV. Genus Iotonchus (Enoplida : Mononchidae). Can. J. Zool., 41 : 79-98.

- MULVEY, R. H. (1963b). The Mononchidae: A family of predaceous nematodes V. Genera Sporonchulus, Granonchulus Prionchuloides n. gen. (Enoplida : Mononchidae). Can. J. Zool., 41 : 763-774.
- MULVEY, R. H. (1967a). The Mononchidae : A family of predaceous nematodes VI. Genus Mononchus (Nematoda : Mononchidae). Can. J. Zool., 45 : 915-940.
- MULVEY, R. H. (1967b). The Mononchidae: A family of predaceous nematodes VII. Genus Prionchulus (Nematoda : Mononchidae). Can. J. Zool., 45 : 941-953.
- MULVEY, R. H. (1978). Predaceous nematodes of the family Mononchidae from MacKenzie and Porcupine river systems and Somerset Island, N.W.T., Canada. Can. J. Zool. 56 : 1849-1860.
- MULVEY, R. H. and JENSEN, H. J. (1967). The Mononchidae of Nigeria. Can. J. Zool., 45 : 667-727.
- NELMES, A. J. (1972). The effects of food deprivation on the hunger of the predaceous soil nematode Prionchulus punctatus. Abstracts of the XIth International Symposium of Nematology, Reading 47.
- NORTON, D. C. (1963). Population fluctuations of Xiphinema americanum in Iowa. Phytopathology, 53 : 66-68.

- O'BANNON, J. H., RADEWALD, J. D. and TOMERLIN, A. T. (1972).  
Population fluctuations of three parasitic nematodes  
in Florida citrus. J. Nematol., 4 : 194-199.
- PAI, S. (1928). Die Phasen des Lebenscyclus der Anquillula  
aceti Ehrbg. und ihre experimentell-morphologische  
Beeinflussung. Z. wiss. Zool., 131 : 293-344.
- PITCHER, R. S. and McNAMARA, D. G. (1972). The toxicity of  
lower concentrations of Silver and Cupric ions to  
three species of plant parasitic nematodes. Nematologica,  
18 : 385-390.
- POTTER, J. M. (1967). Vertical distribution of sting ring  
and stunt nematodes in Norfolk sandy loam following  
peanuts. Nematologica, 13 : 150 (Abtr).
- RASHID, A. and KHAN, A. M. (1978). Morphometric studies on  
Pratylenchus coffeae with description of Pratylenchus  
typicus Rashid, 1974. Indian J. Nematol. 6 : 63-72.
- RASKI, D. J. (1950). The life history and morphology of  
sugarbeet nematode, Heterodera schachtii Schmidt.  
Phytopathology, 40 : 135-152.
- RHODE, R. A. and JENKINS, W. R. (1957). Effect of temperature  
on the life-cycle of stubby-root nematode. Phytopathology,  
47 : 29 (Abstr).
- RICHTER, R. (1969). Zur vertikalen verteilung von Nematodes  
in einen Sandboden. Nematologica, 15 : 44-54.

- RIGGS, R. D., SLACK, D. A. and FULTON, J. P. (1956).  
Meadow nematode and its relation to decline of  
strawberry in Arkansas. Phytopathology, 46 : 24.
- ROBINSON, T. and NEAL, A. L. (1956). The influence of  
hydrogen ion concentration on the emergence of  
golden nematode larvae. Phytopathology, 46 : 665.
- ROMAN, J. and HIRSCHMANN, H. (1969). Embryogenesis and  
Post-embryogenesis in species of Pratylenchus  
(Nematoda : Tylenchidae). Proc. helminth. Soc.  
Wash., 36 : 164-174.
- SINGH, K. and MISRA, S. R. (1968). Spatial distribution of  
plant parasitic nematodes in the root zone of sugar-  
cane. Int. Plant. Path. Congr. (Abstracts) London.
- SOROCZAN, W. (1969). The effect of pH and concentrations  
of mineral salts of the medium on the survival rate  
of larvae and adults of Rhabditis sp. Dujardin under  
laboratory conditions. Annls. Univ. Mariae-Curie.  
Skłodowska, 24 : 153-164.
- STEINER, G. and HEINLEY, H. (1922). The possibility of  
control of Heterodera radicicola and other plant-  
injurious nemas by means of predatory nemas,  
especially by Mononchus papillatus Bastian. J. Wash.  
Acad. Sci., 12 : 367-386.

- STEPHENSON, W. (1945). The effect of Acids on a soil Nematode. Parasitology, 36 : 158-164.
- STURHAN, D. (1963). Allometrische Untersuchungen on Longidoriden. Z. Morph. Ökol. Tiere, 53 : 39-60.
- SZCZYGIEL, A. (1966). Studies on the fauna and population dynamics of nematodes occurring on strawberry plantations. Ekol. Pol. Series A, 14 : 651-709.
- SZCZYGIEL, A. and HASIOR, H. (1972a). Vertical distribution of plant parasitic nematodes in the soil of strawberry plantations. Ekol. Pol. 20 : 493-506.
- SZCZYGIEL, A. and HASIOR, H. (1972b). Seasonal variations in population of plant parasitic nematodes in strawberry plantations. Ekol. Pol. 20 : 507-523.
- TARJAN, A. C. (1964). Two new american dagger nematodes (Xiphinema : Dorylaimidae) associated with citrus, with comments on the variability of X. bakeri Williams, 1961. Proc. helminth. Soc. Wash., 31 : 65-76.
- TARJAN, A. C. (1969). Variation within Xiphinema americanum group (Nematoda : Longidoridae). Nematologica, 15 : 241-252.
- TAYLOR, D. P. and JENKINS, W. R. (1957). Variations within nematode genus Pratylenchus, with description of P. hexincisus n. sp. Nematologica, 2 : 159-174.

- THONG, C. H. S. (1971). Five species of Mononchidae (Nematoda) from Singapore Island. Nematologica, 16 : 470-476.
- THORNE, G. (1927). The life history, habit and economic importance of some mononchs. J. Agric. Res., 37 : 265-286.
- THORNE, G. and MALEK, R. B. (1968). Nematodes of the Northern Great Plains. Tec. Bull. Agric. Exp. Stn. South Dakota state.
- TRIANANTAPHYLLOU, A. C. and HIRSCHMANN, H. (1960). Post-infection development of Meloidogyne incognita Chitwood, 1949 (Nematoda : Heteroderidae). Ann. Inst. Phytopath. Benaki. N. S., 3 : 1-11.
- WEERDT, L. G. VAN. (1958). Studies on the biology of Radopholus similis (Cobb, 1893) Thorne, 1949. Part II. Nematologica, 3 : 184-196.
- WEERDT, L. G. VAN. (1960). Studies on the biology of Radopholus similis (Cobb, 1893) Thorne, 1949. Part III. Embryology and Post-Embryonic Development. Nematologica, 5 : 43-51.
- WALLACE, H. R. (1962). Observations on the behaviour of Ditylenchus disaci in soil. Nematologica, 7 : 91-101.



- WALLACE, H. R. (1963). The Biology of plant parasitic nematodes Edward Arnold Ltd. London : 280 pp.
- WALLACE, H. R. (1971). Abiotic influence in the soil environment. Page 257-280 in B. M. Zuckerman, W. F. Mai, and R. A. Rhode, eds. Plant parasitic nematodes. Vol. 1, Academic Press, New York.
- WALLACE, H. R. and GREET, D. N. (1964). Observations on the taxonomy and biology of Tylenchorhynchus macrurus (Goodey, 1932) Filipjev, 1936, and Tylenchorhynchus icarus sp. nov. Parasitology, 54 : 129-144.
- WU, L. Y. (1960). Comparative study of Ditylenchus destructor Thorne, 1945 (Nematoda : Tylenchidae) from potato, Bulbous iris and dahlia, with discussion of de Man's ratios. Can. J. Zool., 38 : 1175-1188.
- YUEN, P. H. (1965). Further observations on Helicotylenchus vulgaris Yuen. Nematologica, 11 : 623-637.
- YUKSEL, H. S. (1960). Observations on the life cycle of Ditylenchus dipsaci on onion seedlings. Nematologica, 5 : 289-296.
- ZUCKERMAN, B. M., KHERA, S. and PIERCE, A. R. (1964). Population dynamics of nematodes in Cranberry soils. Phytopathology, 54 : 654-659.

TABLE I  
DIMENSIONS OF HADRONCHUS SHAKILI

Populations	N	L mm	a	b	c	c'	V/T	Va	G <sub>1</sub>	G <sub>2</sub>
<u>1. Bareilly population</u>										
♀♀	140	2.40-3.35	35-52	4.4	8.0	7.9	61	67	12	12
					6.4-11.8	5.6-9.5	60-66	65-68	9-15	9-15
♂♂	62	2.32-3.22	35-54	4.4	9.2	5.7	43			
				4.2-5.2	7.3-11.8	4.5-7.0	39-45		-	-
<u>2. Sikkim population</u>										
♀♀	8	1.93-2.12	36-40	4.2	4.9	11.1	56.7	71	12	11
				4.0-4.4	4.7-5.1	10.4-12.0	56-57	70-72	10-15	9-12
♂♂	14	1.79-2.15	36-41	4.2	6.0	7.7	38			
				4.0-4.7	5.4-7.8	5.7-8.4	37-40	-	-	-
<u>3. Malaysia population</u>										
♀♀	2	2.77-2.78	43-45	4.3-4.4	4.6-4.8	13.0-14.0	56-57	70-72	10-12	10

Habitats:

- i) Soil around roots of Litchi, Litchi chinensis from Company Gardens, Bareilly, Uttar Pradesh.
- ii) Soil around roots of orange, Citrus sinensis from Gangtok, Sikkim.
- iii) Soil around roots of Mangosteen tree, Garcinia mangostana, from Sungei Batu, Penang, Malaysia.

TABLE II

## NUMBER OF VULVAL PAPILLAE IN BARCELLO POPULATION

Serial No.	Pre-vulval	Post-vulval	Occurrence
1	0	0	12%
2	1	0	8%
3	0	1	11%
4	1	1	11%
5	2	1	11%
6	1	2	4%
7	2	2	22%
8	3	1	1%
9	3	2	16%
10	2	3	2%
11	0	2	1%
12	4	3	1%

TABLE IV

DIMENSIONS OF JUVENILES OF HADRONCHUS SHAKILLI

Stage	N	L mm	a	b	c	c'
L <sub>1</sub>	16	8.44 (6.12-9.96)	26 (21-31)	3.3 (2.5-3.7)	6.7 (5.5-7.9)	5.9 (5.4-7.3)
L <sub>2</sub>	10	1.26 (1.06-1.45)	32 (29-35)	3.8 (3.4-4.5)	7.1 (6.3-7.7)	6.3 (5.5-7.5)
L <sub>3</sub>	14	1.71 (1.49-1.81)	35 (33-38)	4.1 (3.9-4.2)	8.0 (7.4-8.6)	6.2 (5.6-6.9)
L <sub>4</sub>	22	2.01 (1.81-2.31)	37 (34-43)	4.1 (4.0-4.5)	8.0 (6.6-9.1)	6.8 (5.6-8.5)

Habitat: Soil around roots of Litchi, Litchi chinensis, from  
Company Gardens, Bareilly, Uttar Pradesh.

TABLE V  
BUCCAL CAVITY MEASUREMENTS IN DIFFERENT JUVENILE STAGES, MOULTING SPECIMENS AND ADULTS  
OF HADRONCHUS SHAKILI

	Buccal cavity I	Buccal cavity II	Buccal cavity III
First stage juvenile n = 16	$\bar{X} = 23 \times 10 \text{ um}$ R = 22-24 x 9-11 um		
First moult	23 x 10 um 25 x 10 um 23 x 10 um 22 x 11 um 23 x 11 um	27 x 15 um 29 x 15 um 31 x 13 um 31 x 15 um 31 x 19 um	
Second stage juvenile n = 10		$\bar{X} = 30 \times 10 \text{ um}$ R = 29-31 x 14-17 um	
Second moult		29 x 15 um 29 x 15 um 29 x 15 um 31 x 14 um 28 x 15 um 31 x 17 um	35 x 18 um 35 x 19 um 36 x 19 um 37 x 18 um 37 x 19 um 37 x 23 um

.....: contd :.....

TABLE V: ..... CONTINUED .....

	Buccal cavity III	Buccal cavity IV	Buccal cavity V
Third stage juvenile n = 14	$\bar{X} = 37 \times 19 \text{ um}$ R = 35-39 x 18-20 um		
Third moult	33 x 18 um	40 x 23 um	
	37 x 19 um	46 x 25 um	
	37 x 19 um	46 x 25 um	
	35 x 21 um	46 x 26 um	
	37 x 18 um	48 x 26 um	
	38 x 18 um	48 x 26 um	
	37 x 21 um	49 x 27 um	
Fourth stage juvenile n = 22	$\bar{X} = 45 \times 25 \text{ um}$ R = 41-48 x 22-26 um		
Fourth moult	42 x 25 um	54 x 30 um ( $\delta$ )	
	44 x 26 um	56 x 33 um ( $\delta$ )	
	46 x 26 um	60 x 36 um ( $\sigma$ )	
Adults ( $\sigma\sigma$ ) n=140		$\bar{X} = 60 \times 36 \text{ um}$ R = 50-65 x 32-40 um	
Adults ( $\sigma\sigma$ ) n=62		$\bar{X} = 55 \times 31 \text{ um}$ R = 49-66 x 29-35 um	

 $\bar{X}$  = Mean      R = Range

TABLE VI  
SURVIVAL TIMES OF ADULTS AND JUVENILES OF HADRONCHUS SHAKILI

pH of the phosphate buffer	Female		Male		L <sub>4</sub>		L <sub>3</sub>		L <sub>2</sub>		L <sub>1</sub>	
	Buffer Solu- tion (sec)	Tap Water (min)	Buffer Solu- tion (sec)	Tap Water (min)	Buffer Solu- tion (sec)	Tap Water (min)	Buffer Solu- tion (sec)	Tap Water (min)	Buffer Solu- tion (sec)	Tap Water (min)	Buffer Solu- tion (sec)	Tap Water (min)
2.2	150	-	110	-	93	-	70	-	60	-	40	-
3.0	410	-	370	-	340	-	240	-	220	-	150	-
4.0	430	761	375	750	370	720	243	310	240	300	160	-
5.0	428	2400	390	2160	370	2160	250	330	240	300	160	120
5.4	440	2400	389	2210	380	2340	270	540	241	450	170	120
5.6	450	2580	430	2460	390	2400	267	690	260	470	170	430
5.8	600	3660	570	3480	530	3120	360	810	340	800	250	560
6.0	490	3420	480	3260	450	3040	300	780	300	730	200	540
6.4	490	1740	470	1560	440	1440	300	690	290	590	200	330
7.0	480	1380	380	1430	370	1380	240	670	240	570	160	310
7.4	410	1180	330	1160	330	1080	210	560	200	380	130	320
8.0	320	1150	250	980	240	960	160	540	160	360	110	320

TABLE VII  
SURVIVAL TIME OF ADULTS AND JUVENILES OF HADRONCHUS SHAKILI  
IN MINERAL SALTS

Mineral salt concentrations	Survival time					
	Female	Male	L <sub>4</sub>	L <sub>3</sub>	L <sub>2</sub>	L <sub>1</sub>
Potassium carbonate						
0.4 M	10	7	7	5	3	3
0.3 M	11	9	9	9	5	5
0.2 M	40	36	36	29	20	12
0.1 M	109	104	95	47	35	27
0.05 M	151	147	133	112	63	53
Potassium chloride						
0.4M	3	3	2	2	1	1
0.3 M	5	4	4	3	2	2
0.2 M	18	15	14	10	9	6
0.1 M	67	64	56	42	38	27
0.05 M	129	121	117	84	70	42
Potassium nitrate						
0.4 M	3	3	3	2	1	1
0.3 M	4	3	3	2	1	1
0.2 M	10	8	7	4	2	2
0.1 M	17	14	11	8	6	4
0.05 M	37	33	27	19	16	9

.....: contd :.....



TABLE VII: .... CONTINUED .....

Mineral salt concentrations	Survival time					
	Female	Male	L <sub>4</sub>	L <sub>3</sub>	L <sub>2</sub>	L <sub>1</sub>
Copper sulphate						
0.4 M	2	2	2	1	1	37 sec.
0.3 M	2	2	2	1	1	1
0.2 M	4	3	3	2	2	1
0.1 M	11	10	7	6	4	3
0.05 M	15	15	13	9	8	5

All readings are in min (=Minutes) unless otherwise stated.

TABLE VIII  
SURVIVAL TIME OF ADULTS AND JUVENILES OF HADRONCHUS SHAKILI IN  
FATTY ACIDS

Fatty acid concentrations	Female	Male	Survival time			
			L <sub>4</sub>	L <sub>3</sub>	L <sub>2</sub>	L <sub>1</sub>
Acetic acid						
1.0 N	4	3	3	2	2	1
0.8 N	5	5	4	4	3	2
0.6 N	8	6	5	4	4	2
0.4 N	9	9	8	5	5	3
0.2 N	12	11	11	7	6	4
0.1 N	16	15	12	9	9	5
0.01 N	51	47	45	39	32	19
0.001 N	1695	1633	1580	1300	1259	785
Propionic acid						
1.0 N	4	3	3	2	2	1
0.8 N	4	4	4	2	2	1
0.6 N	8	8	6	3	3	1
0.4 N	10	10	9	5	4	2
0.2 N	10	10	10	7	6	3
0.1 N	15	13	13	10	9	3
0.01 N	43	36	34	31	20	14
0.001 N	1604	1591	1559	1187	1167	765

.....: contd :.....

TABLE VIII: ..... CONTINUED .....

Fatty acid concentrations	Female	Male	Survival time			
			L <sub>4</sub>	L <sub>3</sub>	L <sub>2</sub>	L <sub>1</sub>
Butyric acid						
1.0 N	3	3	3	2	2	19 sec
0.8 N	5	4	3	2	2	27 sec
0.6 N	5	5	4	3	3	37 sec
0.4 N	6	5	5	5	4	56 sec
0.2 N	7	7	6	5	4	1
0.1 N	14	13	13	9	7	2
0.01 N	37	33	31	23	20	11
0.001 N	1598	1583	1552	1173	1140	627
Formic acid						
1.0 N	1	34 sec	25 sec	21 sec	16 sec	12 sec
0.8 N	1	1	33 sec	29 sec	28 sec	22 sec
0.6 N	1	1	1	37 sec	30 sec	28 sec
0.4 N	2	2	1	1	1	1
0.2 N	3	3	3	2	2	1
0.1 N	3	3	3	2	2	2
0.01 N	9	8	8	7	6	4
0.001 N	1373	1327	1300	1079	1028	523

All readings are in min (= minutes) unless otherwise stated.

Fig. I

- A - Entire female,
- B - Entire male,
- C - Buccal cavity (Lateral view),
- D - Buccal cavity (Dorso-ventral view),
- E - Oesophagus.

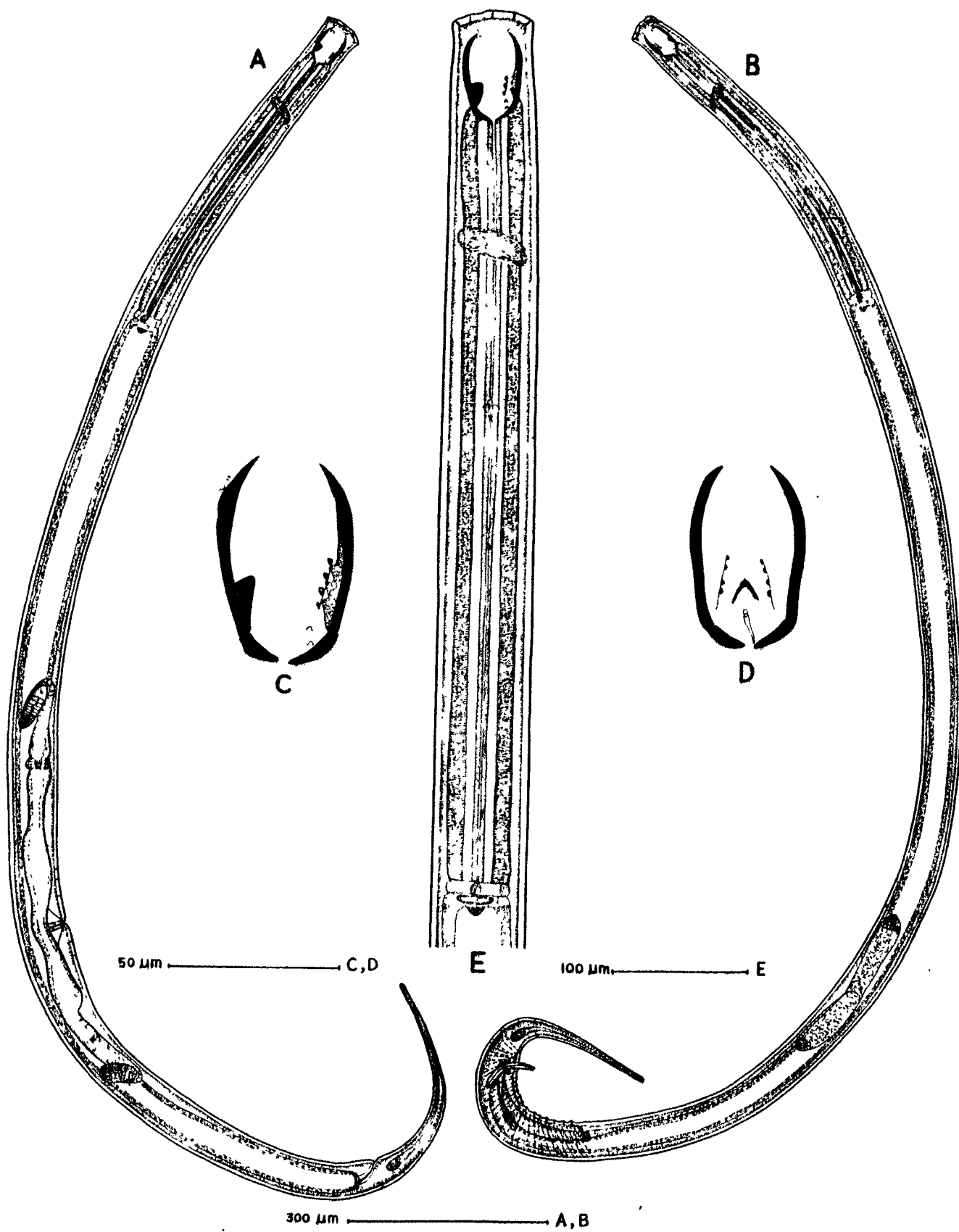
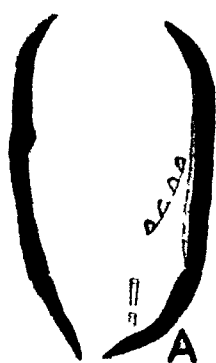


Fig. II

- A-C - Abnormal buccal cavities,
- D - Anterior region of body showing a nematode (prey)  
in the buccal cavity.
- E-G - Abnormal tails.
- H - Vagina without cuticularized pieces.
- I - Posterior region of male showing abnormal number  
of ejaculatory glands (five).



A



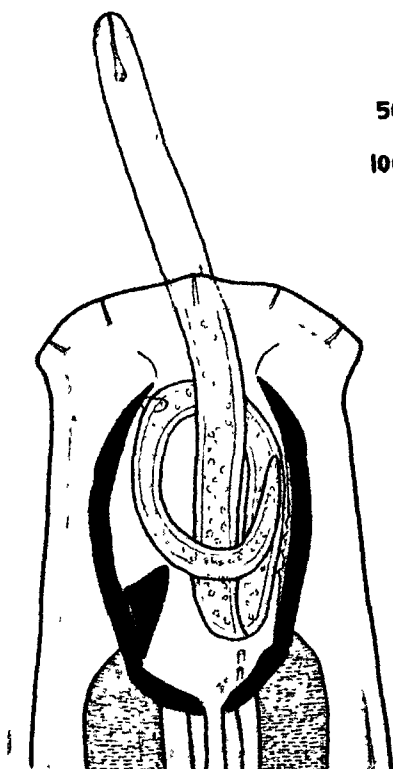
B



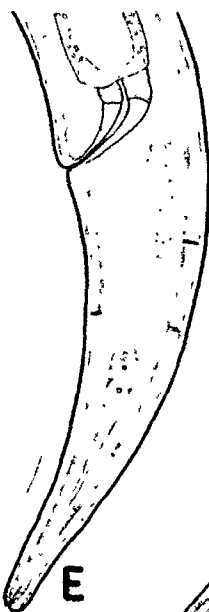
C

50  $\mu$ m  $\longleftrightarrow$  A-D, H

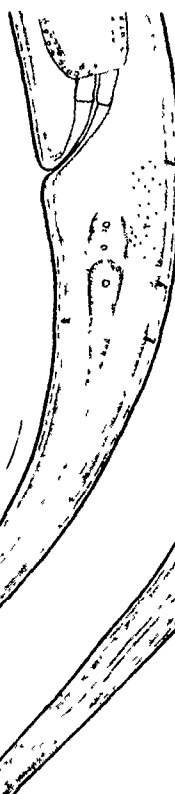
100  $\mu$ m  $\longleftrightarrow$  E-G, I



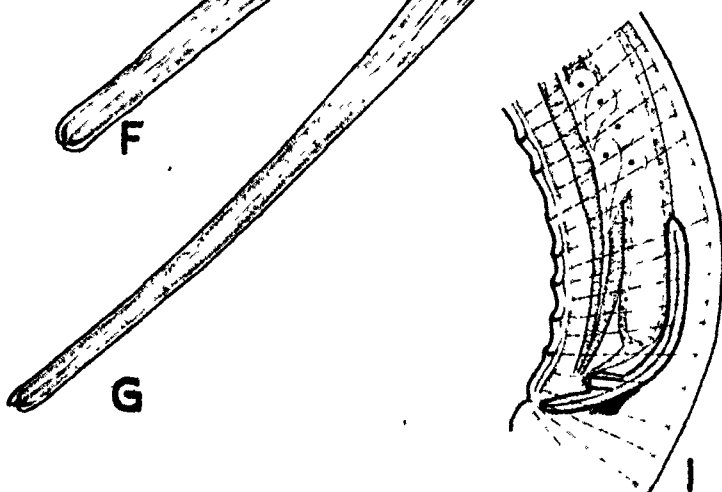
D



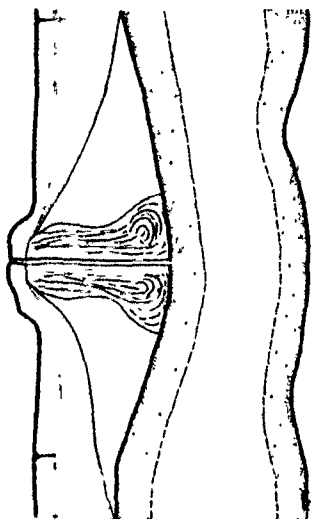
E



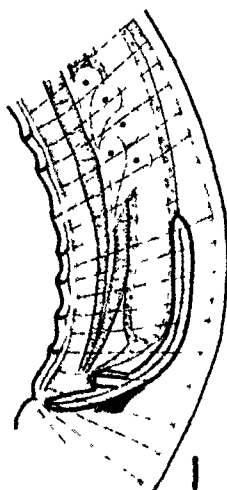
F



G



H



I

Fig. III

Variation in number and position of the vulval papillae.



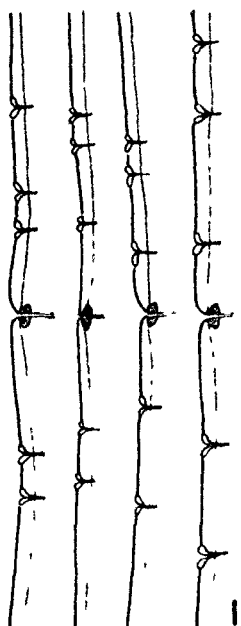
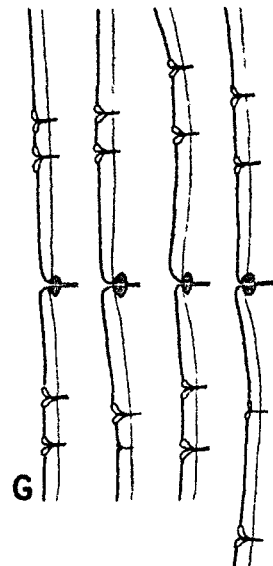
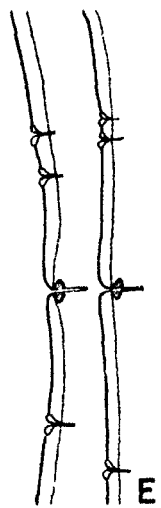
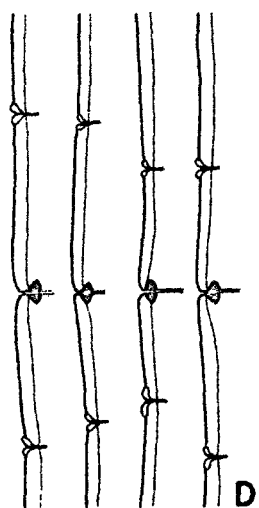
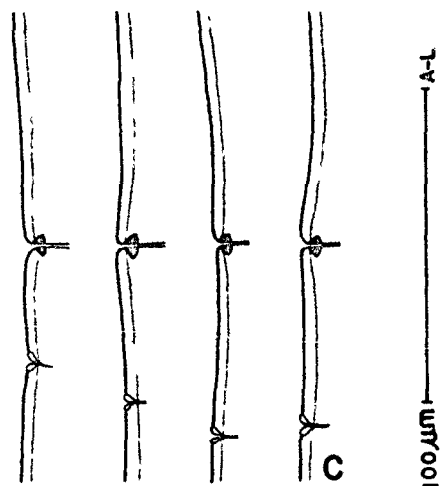
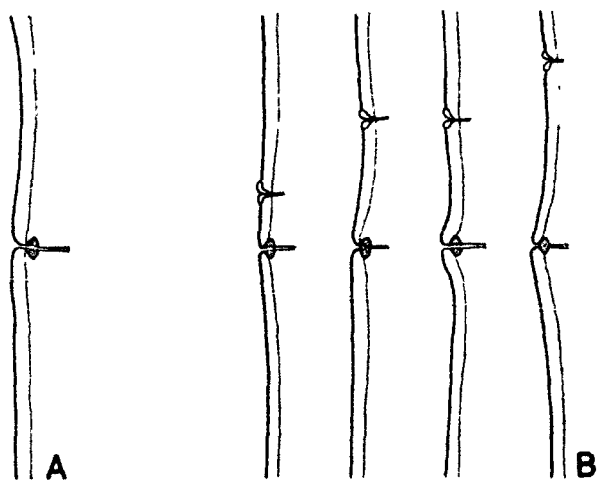


Fig. IV

Female anterior sexual branches.

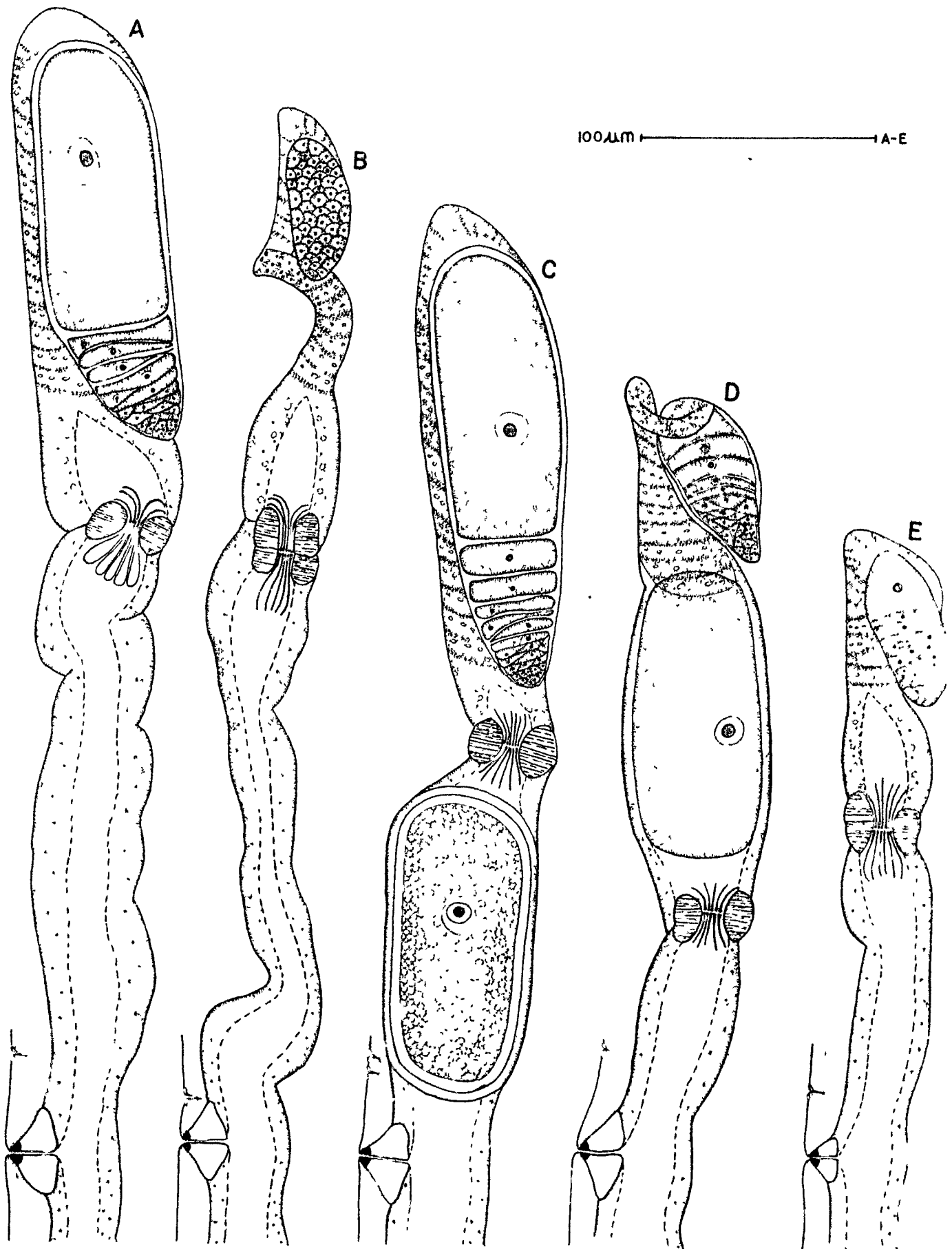


Fig. V

Female posterior sexual branches

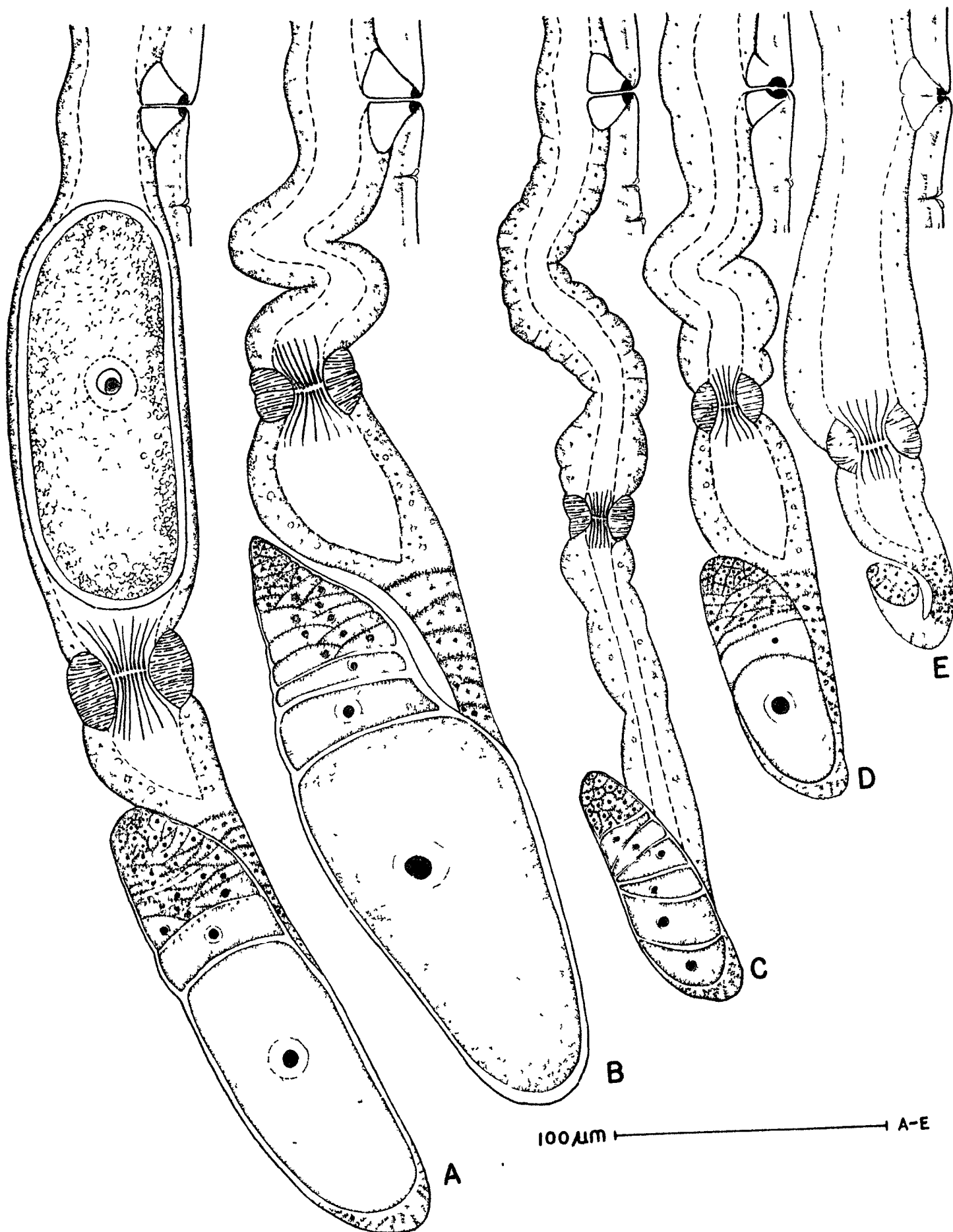
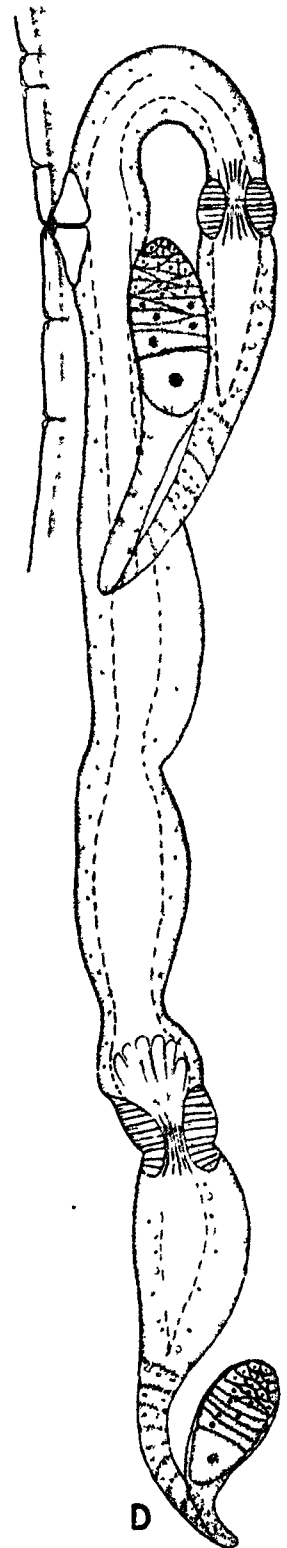
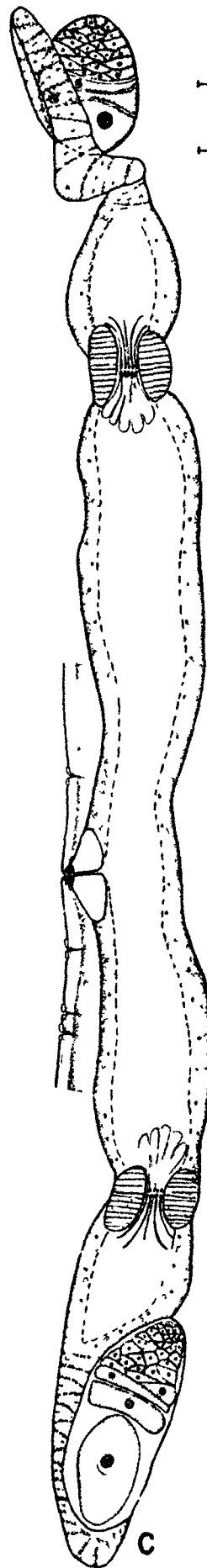
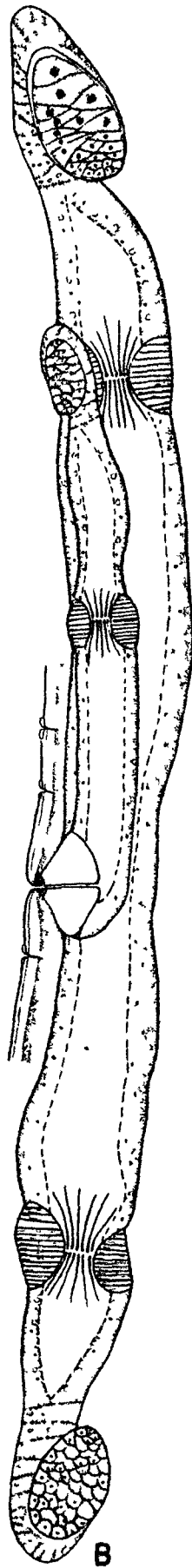
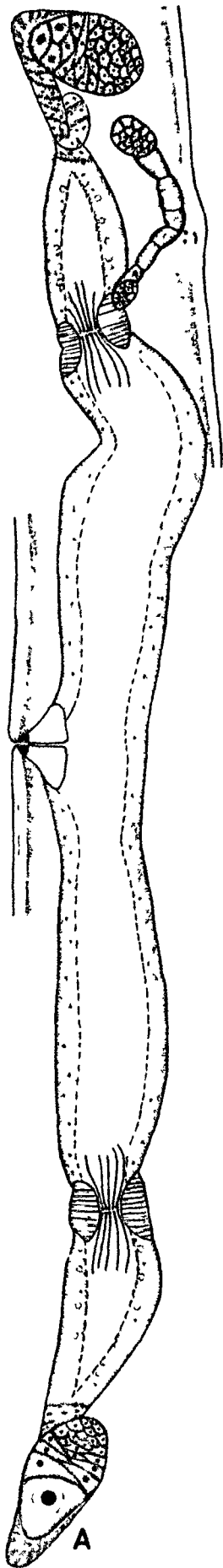


Fig. VI

Abnormal female gonads.



100  $\mu$ m A,C

100  $\mu$ m B,D

Fig. VII

Female tails: A & B - Bareilly population;  
C & D - Sikkim population;  
E & F - Malaysia population.



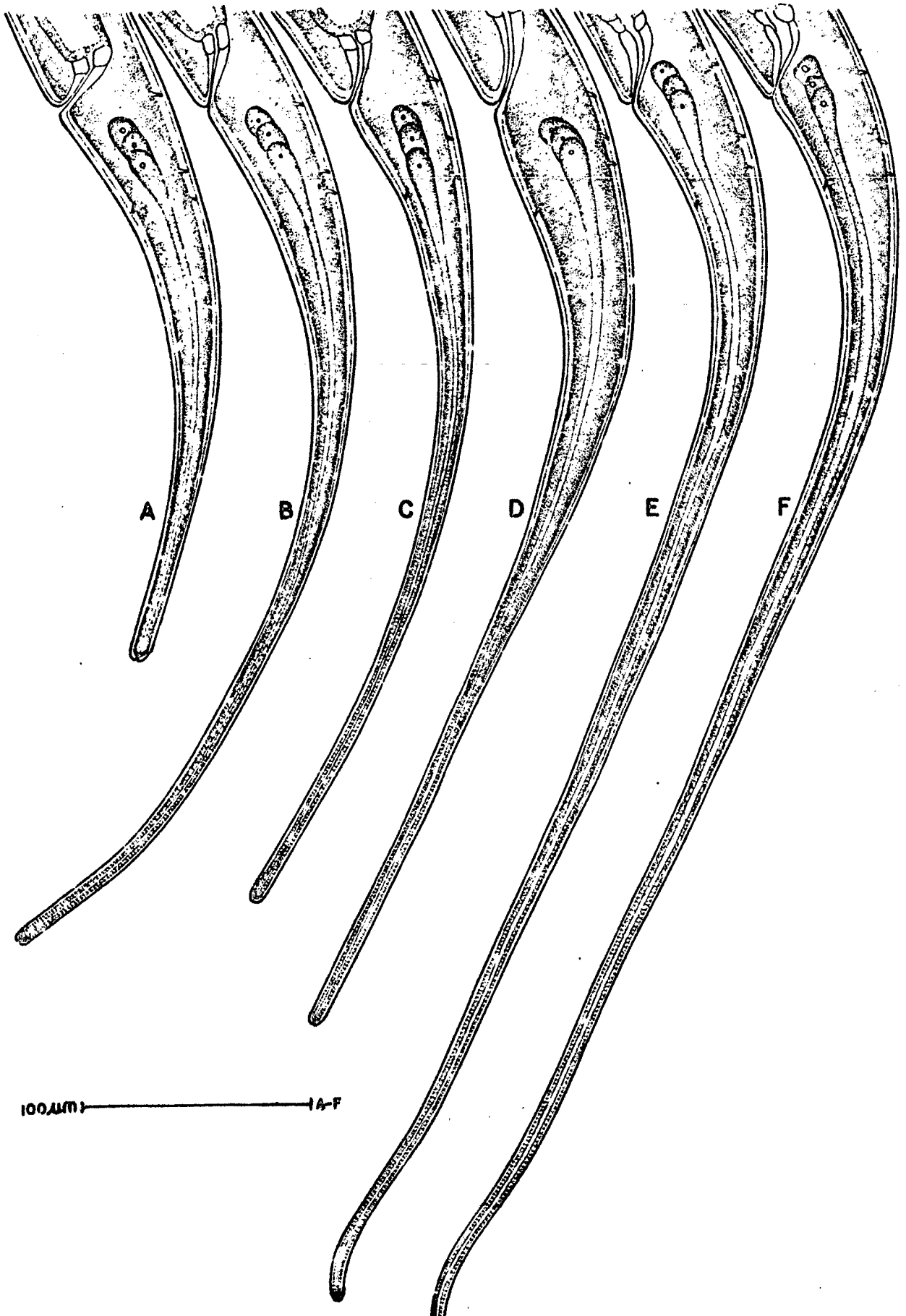


Fig. VIII

Male tails: A - E Bareilly population;  
F & G Sikkim population.

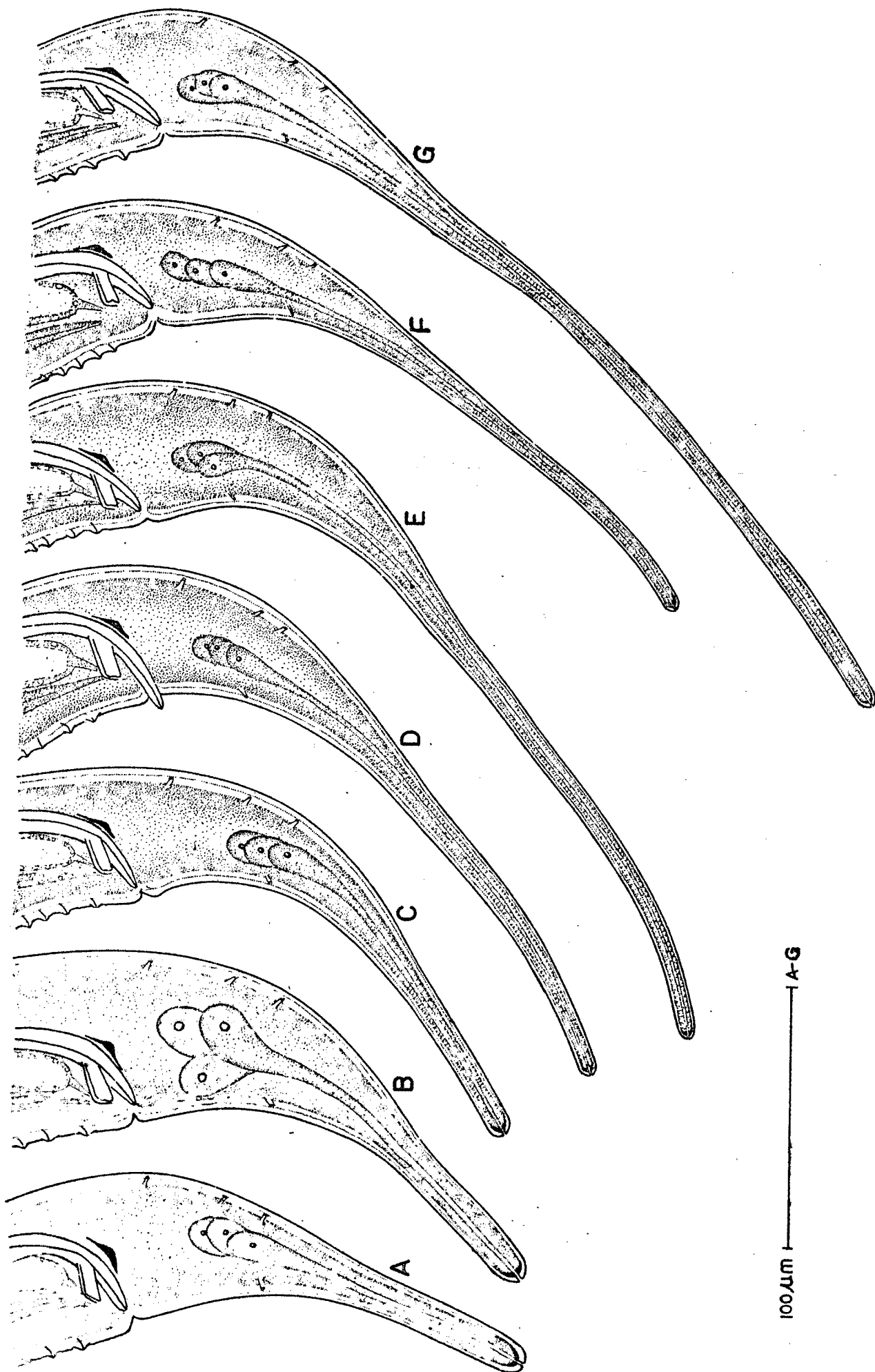


Fig. IX

## MUSCULATURE

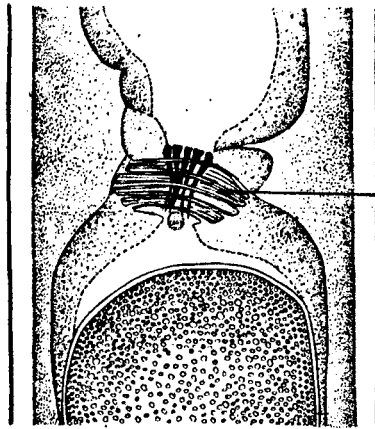
Specialized muscles

- A - Sphincter muscles,
- B - Cephalic muscles,
- C & D - Vulval muscles,
- E - Anal muscles (female),
- G - Spicular muscles,
- H - Anal, copulatory, gubernacular and caudal copulatory muscles (male).

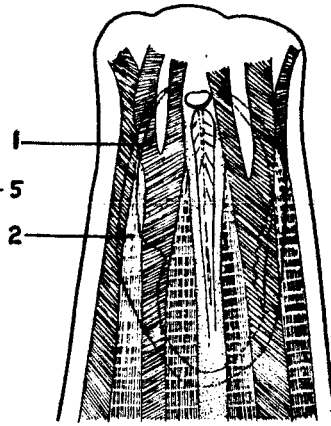
Somatic muscles (Cross-sections)

- I - At base of buccal cavity
- J - At level of nerve ring
- K - At level of anus.

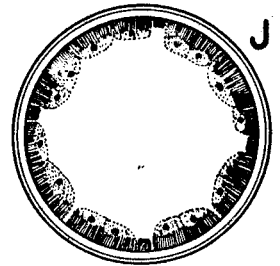
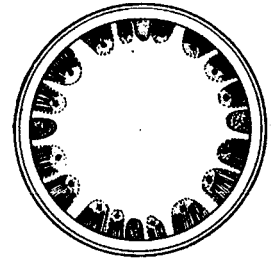
1. Labial muscles, 2. Stomatal muscles, 3. Dilator muscles, 4. Constrictor vulvae, 5. Sphincter muscles, 6. Anal muscles, 7. Copulatory muscles, 8. Accessory copulatory muscles, 9. Retractor gubernaculi, 10. Retractor spiculi, 11. Protractor spiculi, 12. Caudal copulatory muscles.



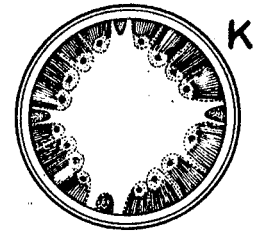
A



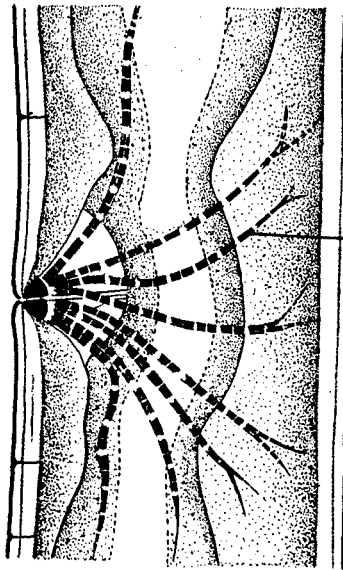
B



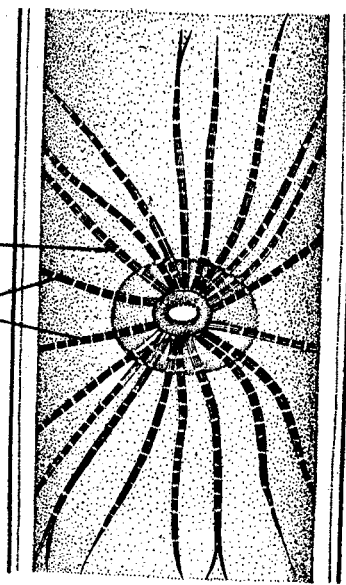
J



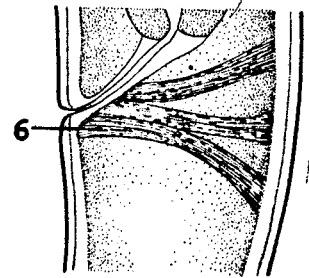
K



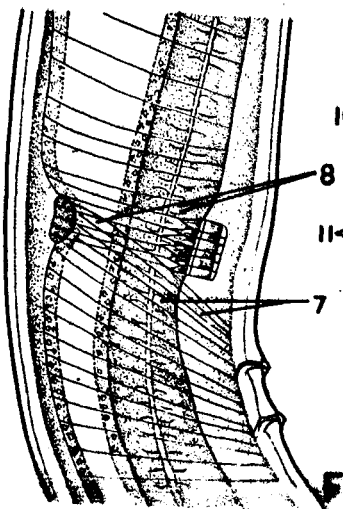
C



D



E

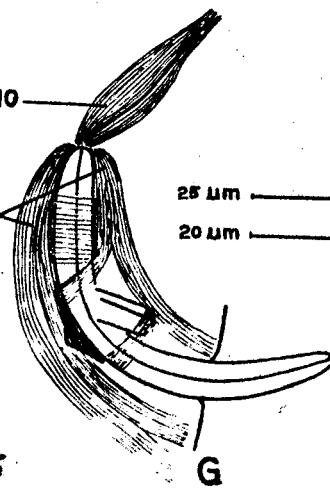


10

8

7

F



10

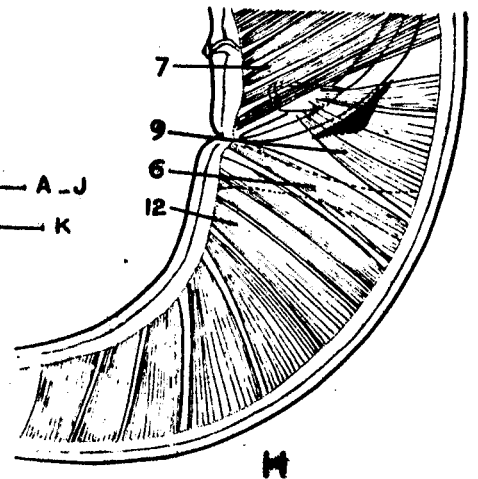
8

11

G

25  $\mu$ m — A-J

20  $\mu$ m — K



7

9

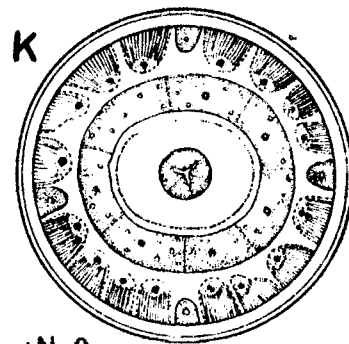
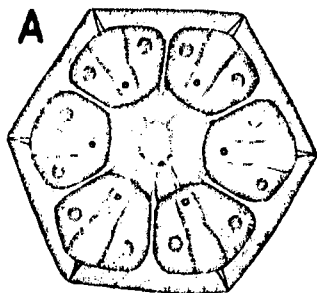
6

12

H

Fig. X  
CROSS-SECTIONS OF BODY

- A - En face view,
- B - Above dorsal tooth,
- C - At level of dorsal tooth,
- D - At basal level of dorsal tooth showing cephalic muscles,
- E - At level of formina,
- F - At base of buccal cavity,
- G - At level of nerve ring,
- H - Through middle of oesophagus,
- I - At base of oesophagus,
- J - Through tubercles,
- K - Through conical organ of oesophago-intestinal junction,
- L - Through intestine,
- M - Through rectum,
- N - At level of anus,
- O - At level of caudal glands.



25  $\mu$ m — A-M

20  $\mu$ m — N-O

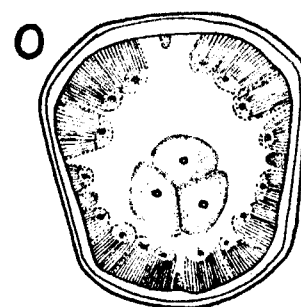
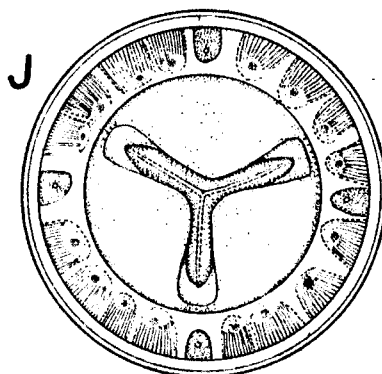
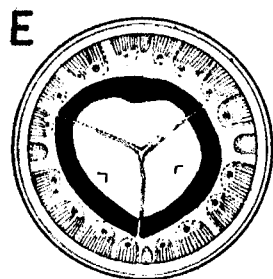
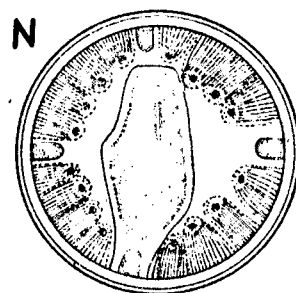
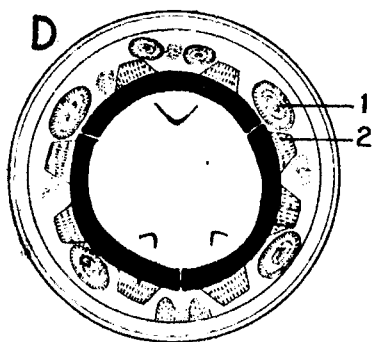
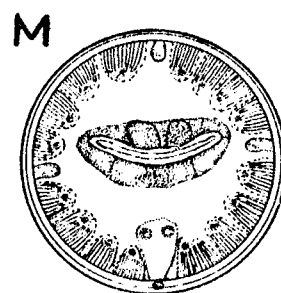
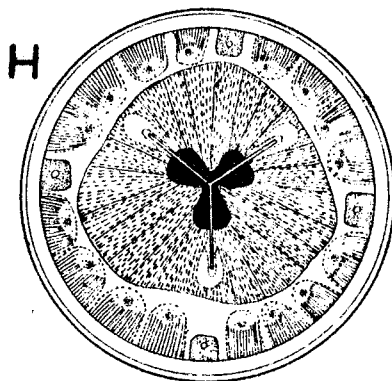
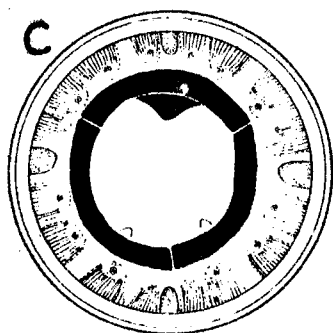
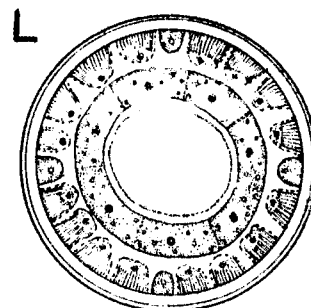
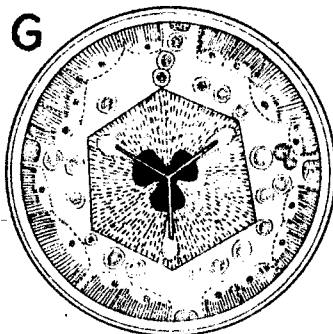
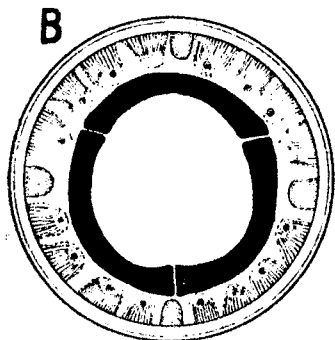
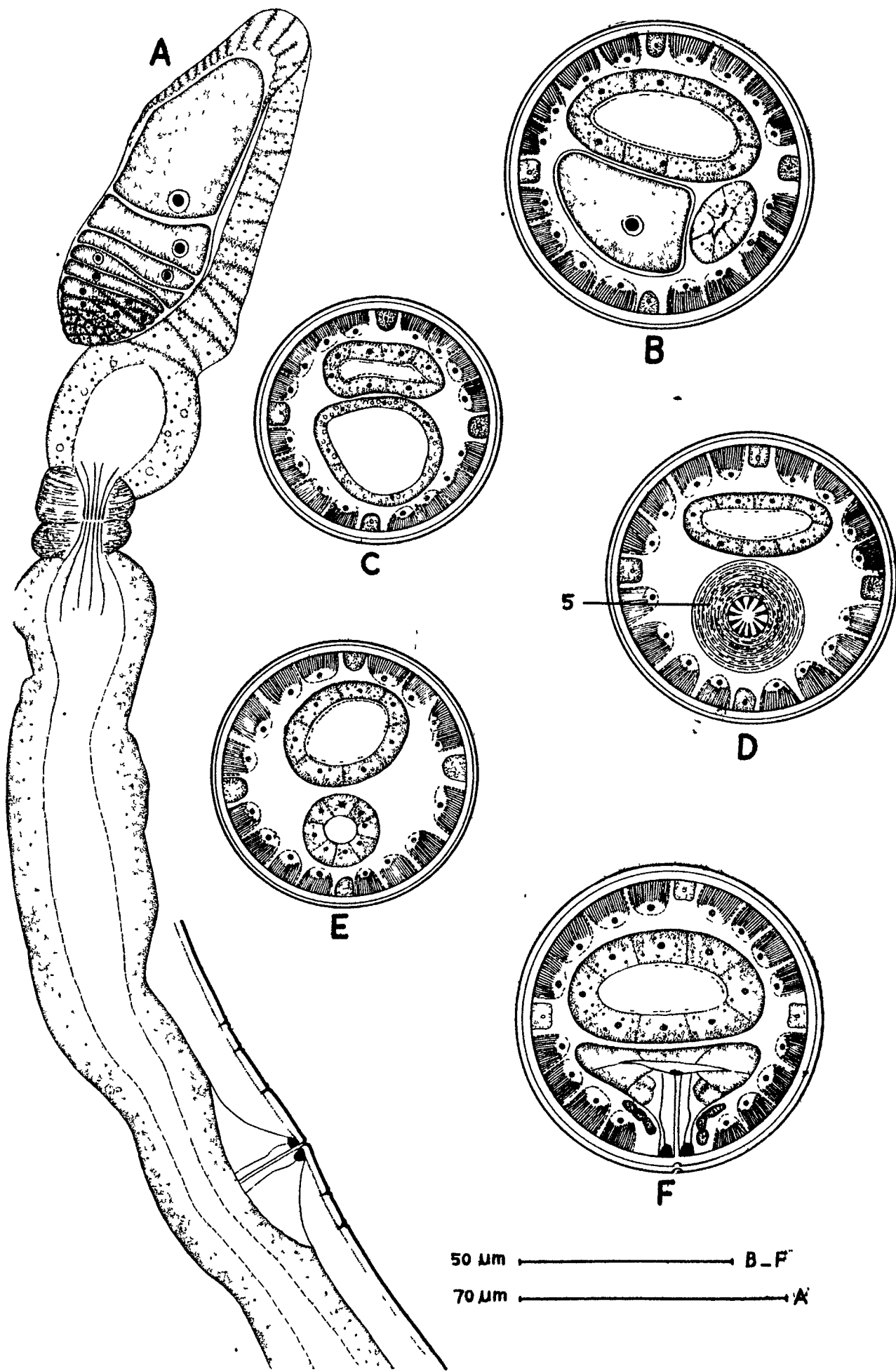


Fig. XI  
FEMALE GONAD

- A     -     Anterior gonad,
- B     -     C. S. through ovary and distal part of oviduct,
- C     -     C. S. through proximal part of oviduct,
- D     -     C. S. at level of sphincter,
- E     -     C. S. through uterus,
- F     -     C. S. at level of vulva.

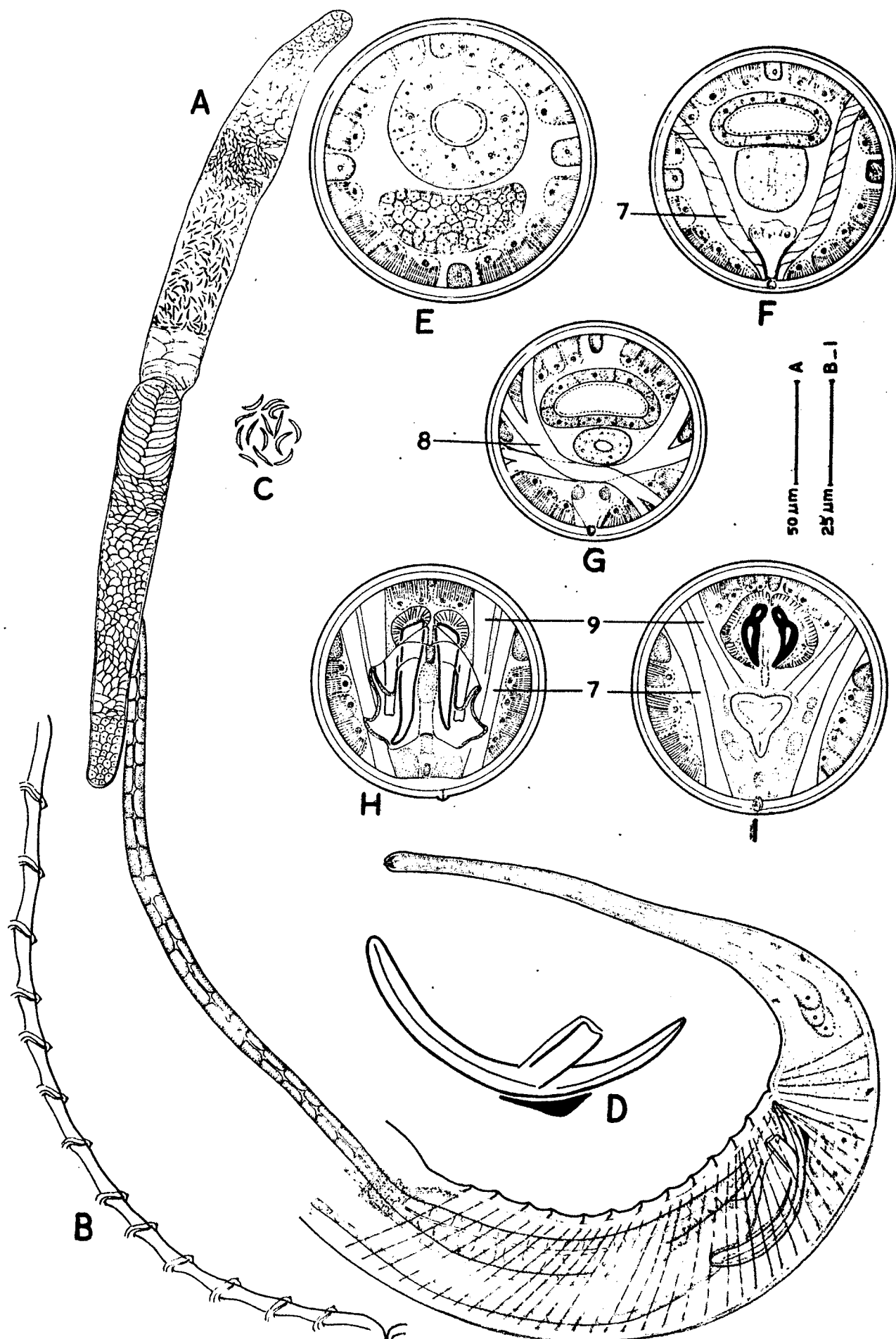




## Fig. XII

## MALE GONAD

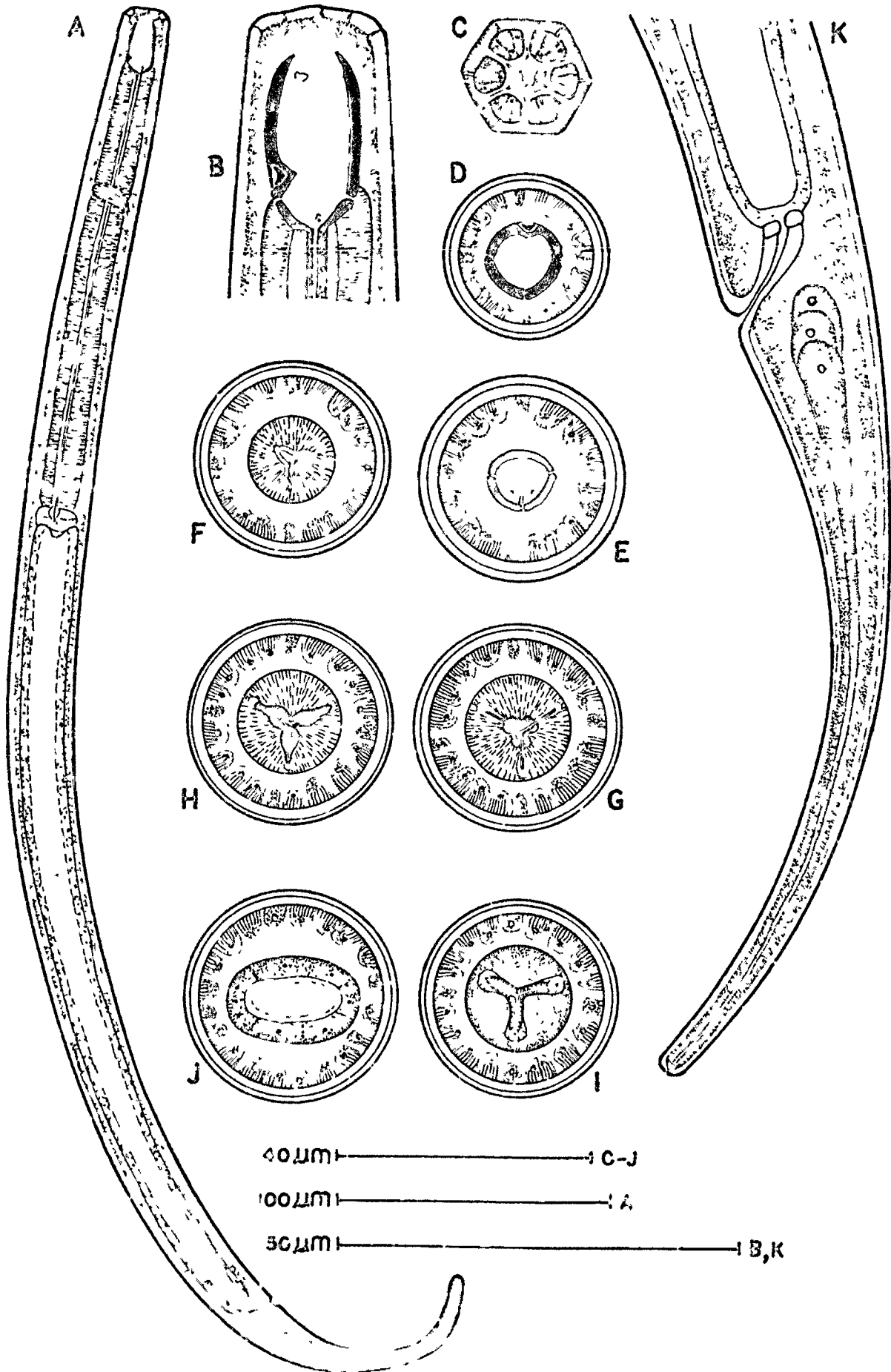
- |   |   |   |
|---|---|---|
| A | - | Entire gonad,   |
| B | - | Supplements,  |
| C | - | Sperms,   |
| D | - | Spicule, gubernaculum and accessory pieces,                             |
| E | - | C. S. through germinal zone of testis,                                  |
| F | - | C. S. through vas deferens,   |
| G | - | C. S. at level of transversly oriented accessory<br>copulatory muscles, |
| H | - | C. S. through spicule,  |
| I | - | C. S. through head of spicule.  |



## Fig. XIII

## FIRST STAGE JUVENILE

- |   |   |                                     |
|---|---|-------------------------------------|
| A | - | Entire,                             |
| B | - | Head end,                           |
| C | - | <u>En face</u> view,                |
| D | - | C. S. at level of dorsal tooth,     |
| E | - | C. S. at level of foramina,         |
| F | - | C. S. at base of buccal cavity,     |
| G | - | C. S. through middle of oesophagus, |
| H | - | C. S. at base of oesophagus,        |
| I | - | C. S. through tubercles,            |
| J | - | C. S. through intestine,            |
| K | - | fail.                               |



40 μm ————— C-J

100 μm ————— A

30 μm ————— B, K

## Fig. XIV

## SECOND STAGE JUVENILE

- |   |   |                                     |
|---|---|-------------------------------------|
| A | - | Entire,                             |
| B | - | Head end,                           |
| C | - | <u>En face</u> view,                |
| D | - | C. S. at level of dorsal tooth,     |
| E | - | C. S. at level of foramina,         |
| F | - | C. S. at base of buccal cavity,     |
| G | - | C. S. through middle of oesophagus, |
| H | - | C. S. at base of oesophagus,        |
| I | - | C. S. through tubercles,            |
| J | - | C. S. through intestine,            |
| K | - | Tail.                               |

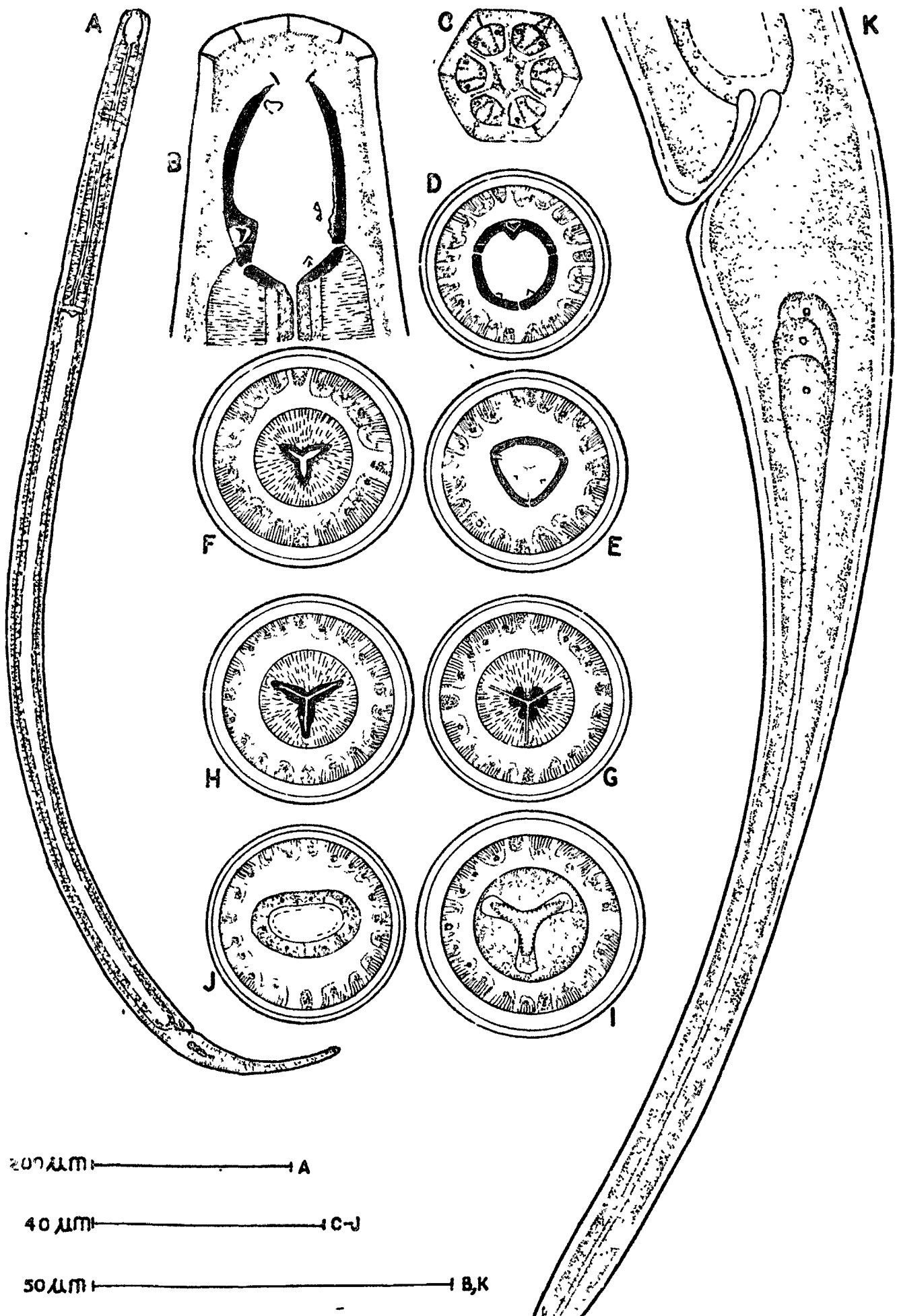


Fig. XV

## THIRD STAGE JUVENILE

- |    |   |                                     |
|----|---|-------------------------------------|
| A  | - | Entire,                             |
| B  | - | Head end,                           |
| C  | - | <u>En face</u> view,                |
| D  | - | C. S. at level of dorsal tooth,     |
| E  | - | C. S. at level of foramina,         |
| F. | - | C. S. at base of buccal cavity,     |
| G  | - | C. S. through middle of oesophagus, |
| H  | - | C. S. at base of oesophagus,        |
| I  | - | C. S. through tubercles,            |
| J  | - | C. S. through intestine,            |
| K  | - | Tail.                               |



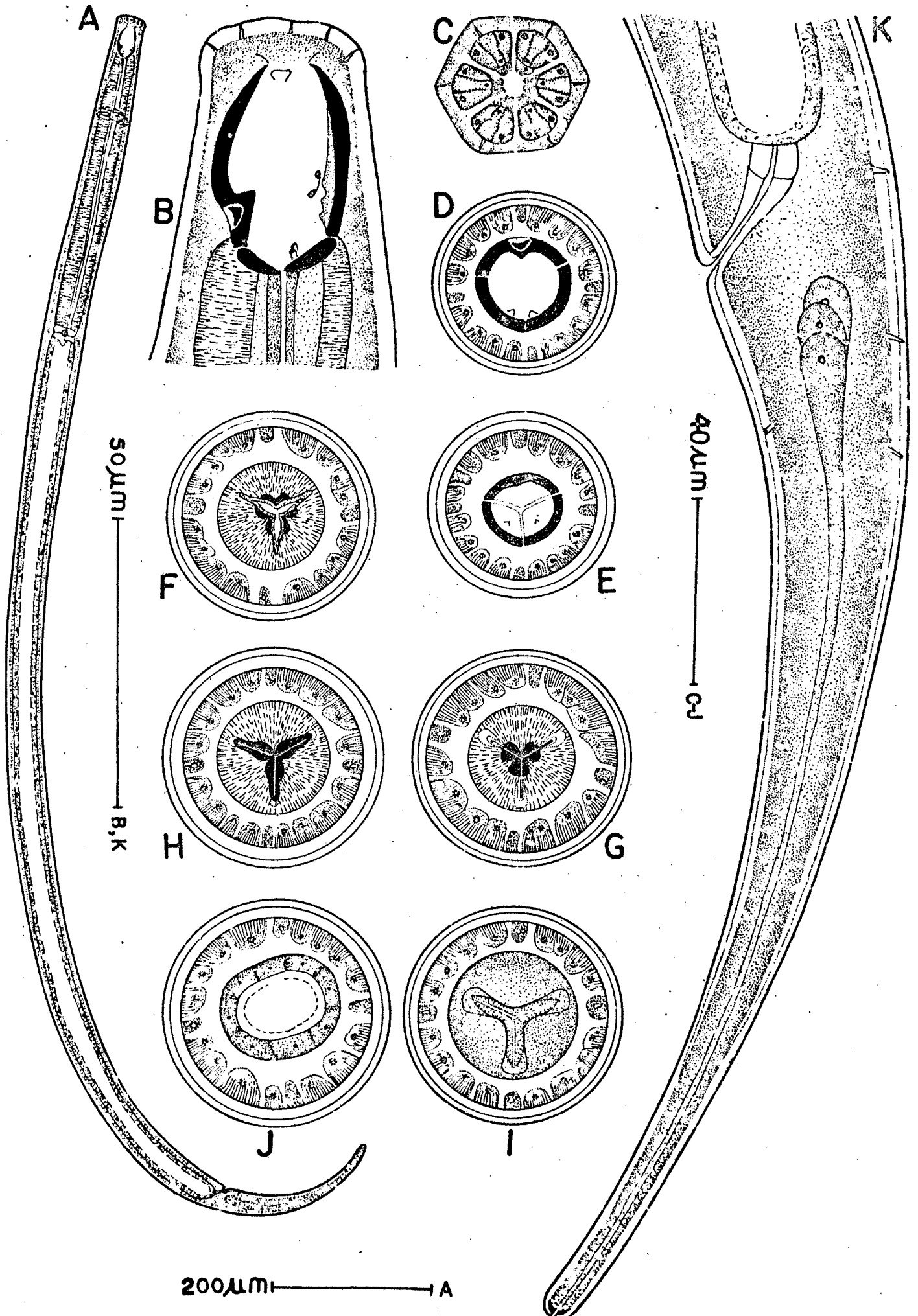


Fig. XVI

## FOURTH STAGE JUVENILE

- |   |   |                                     |
|---|---|-------------------------------------|
| A | - | Entire,                             |
| B | - | Head end,                           |
| C | - | <u>En face</u> view,                |
| D | - | C. S. at level of dorsal tooth,     |
| E | - | C. S. at level of foramina,         |
| F | - | C. S. at base of buccal cavity,     |
| G | - | C. S. through middle of oesophagus, |
| H | - | C. S. at base of oesophagus,        |
| I | - | C. S. through tubercles,            |
| J | - | C. S. through intestine,            |
| K | - | Tail.                               |

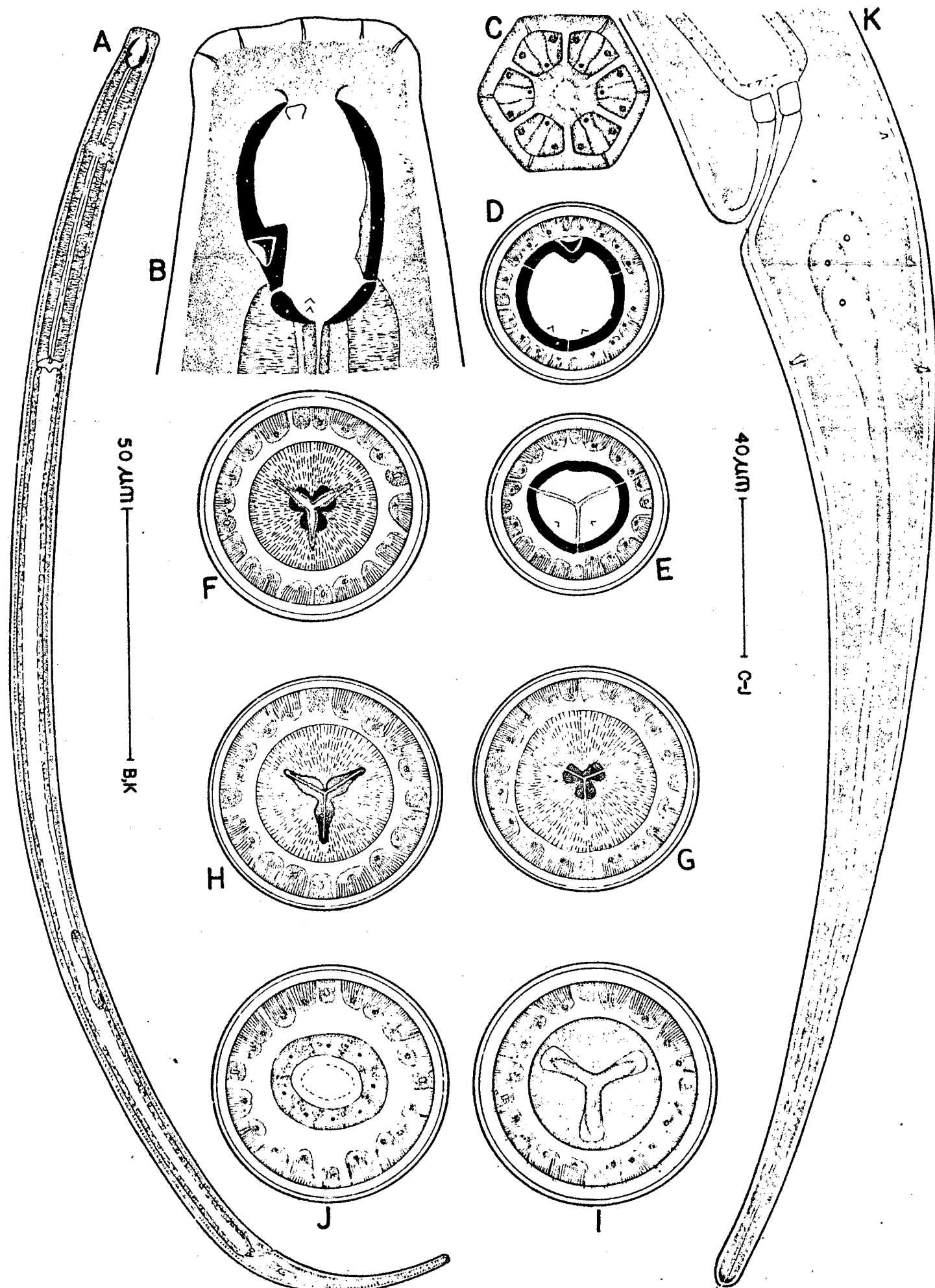


Fig. XVII

The body length in relation to A - buccal cavity length;  
B - oesophageal length; C - tail length in adults.

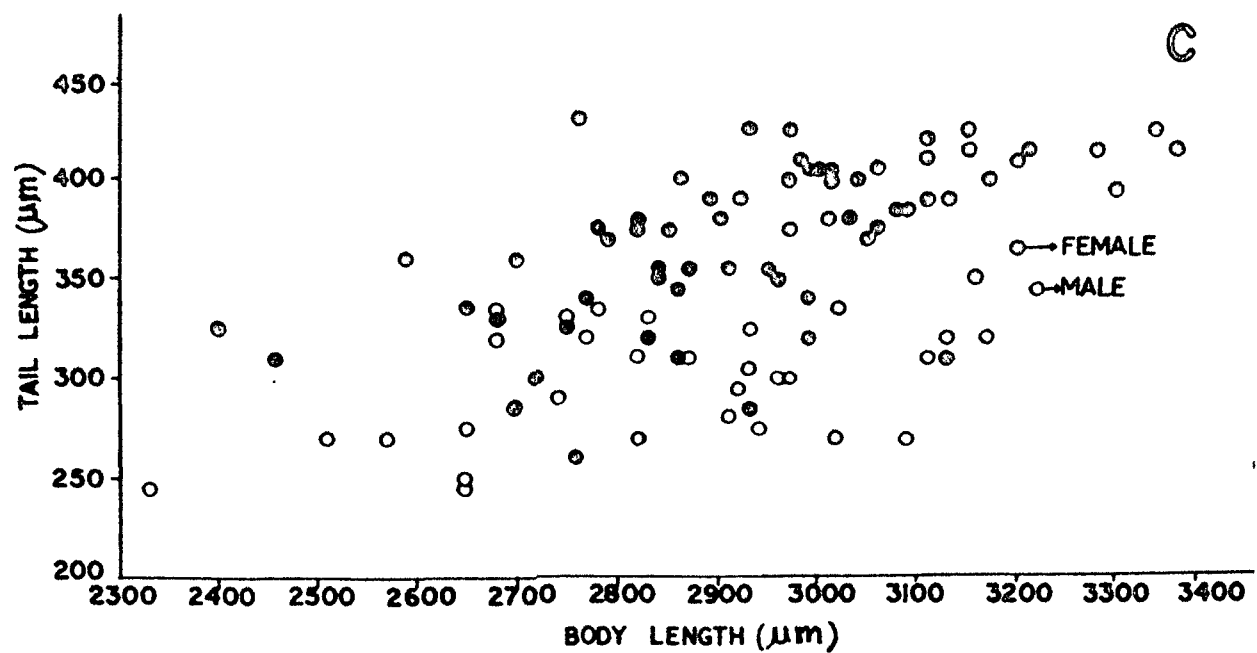
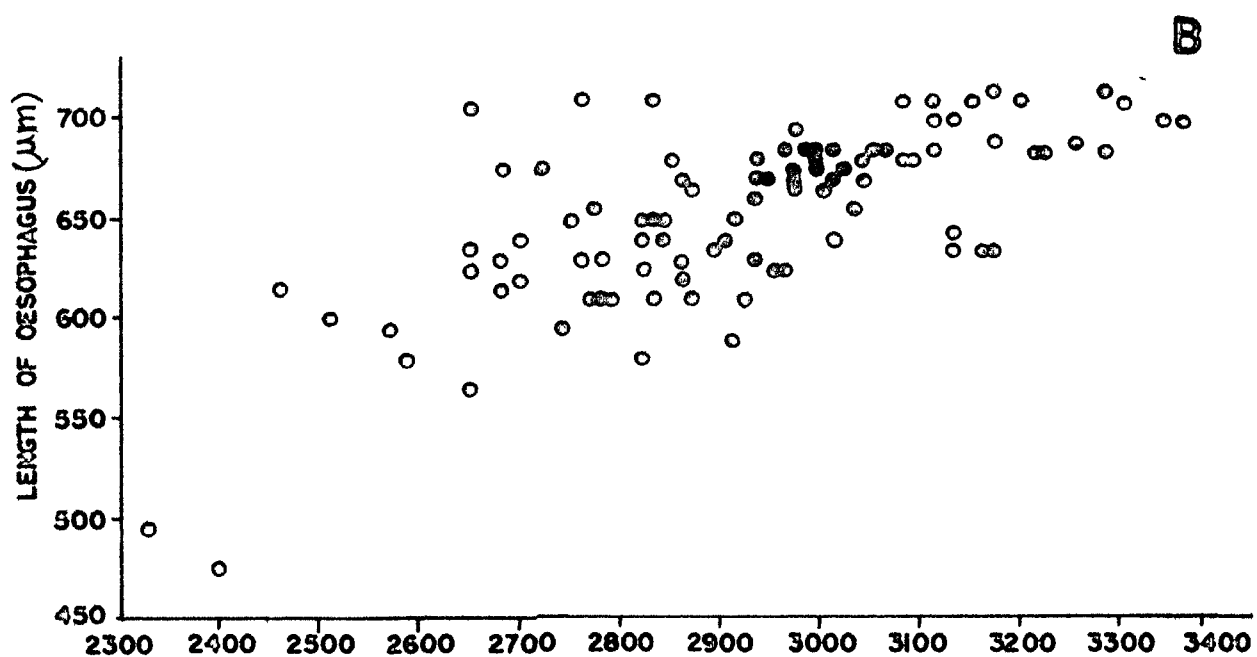
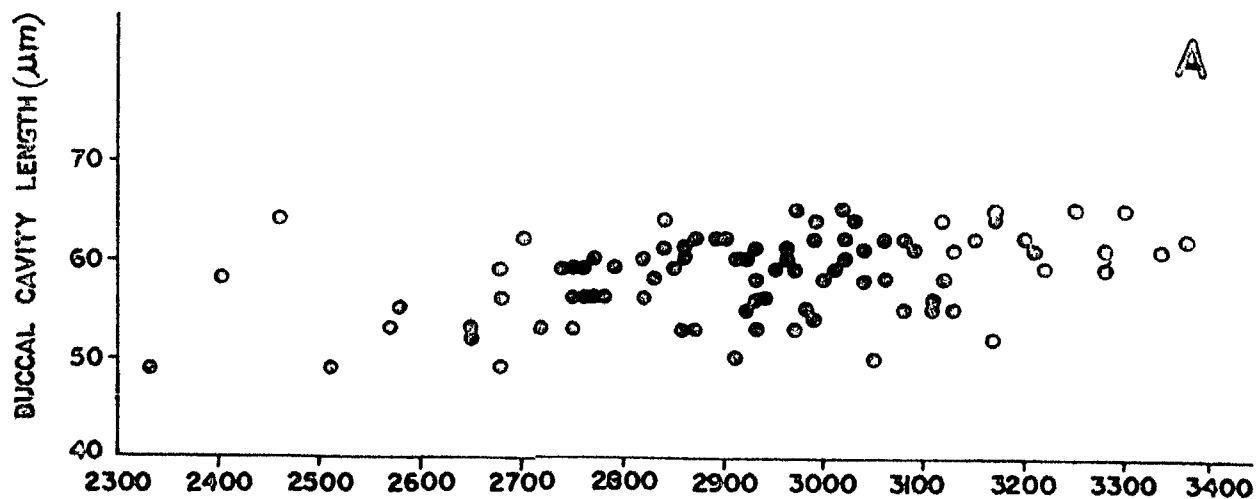


Fig. XVIII

The body length in relation to A - vulva position, B - gonad length.

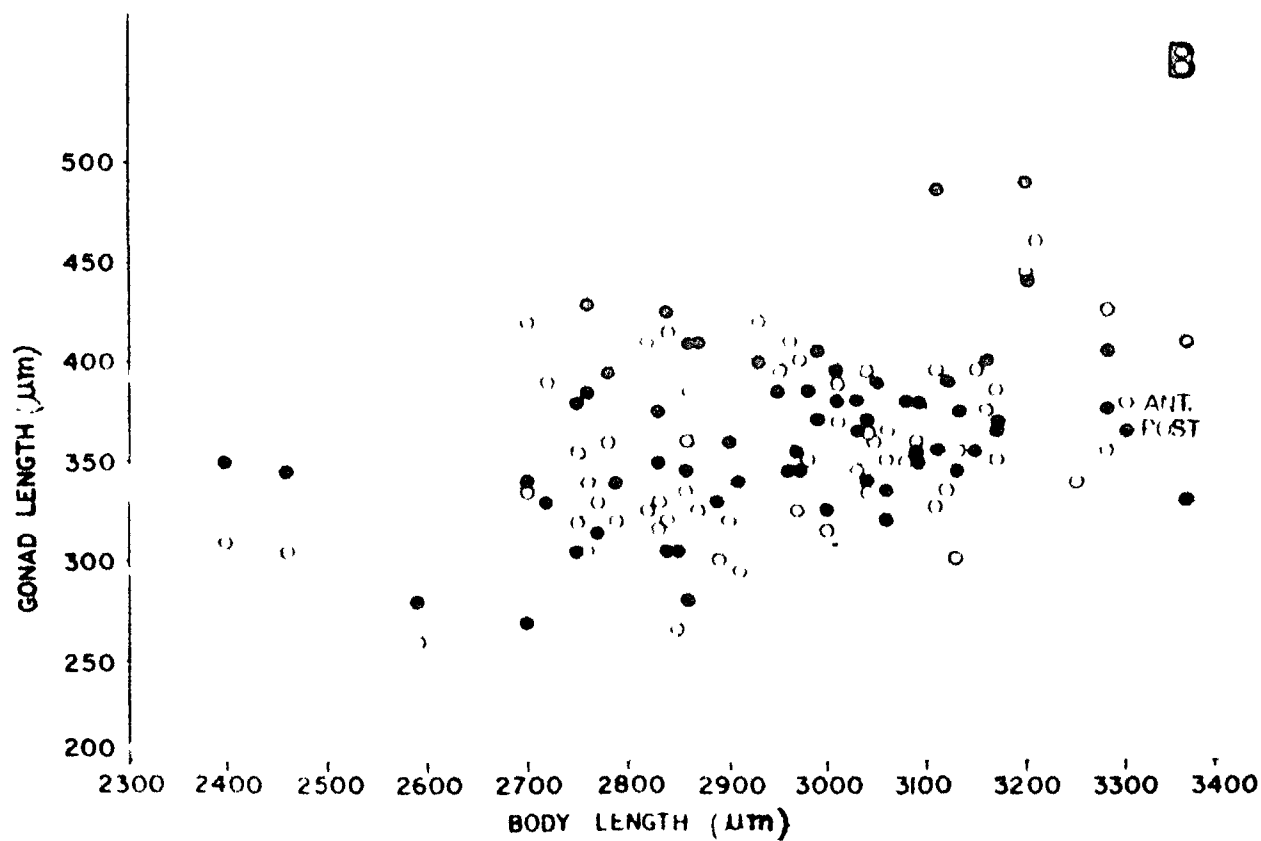
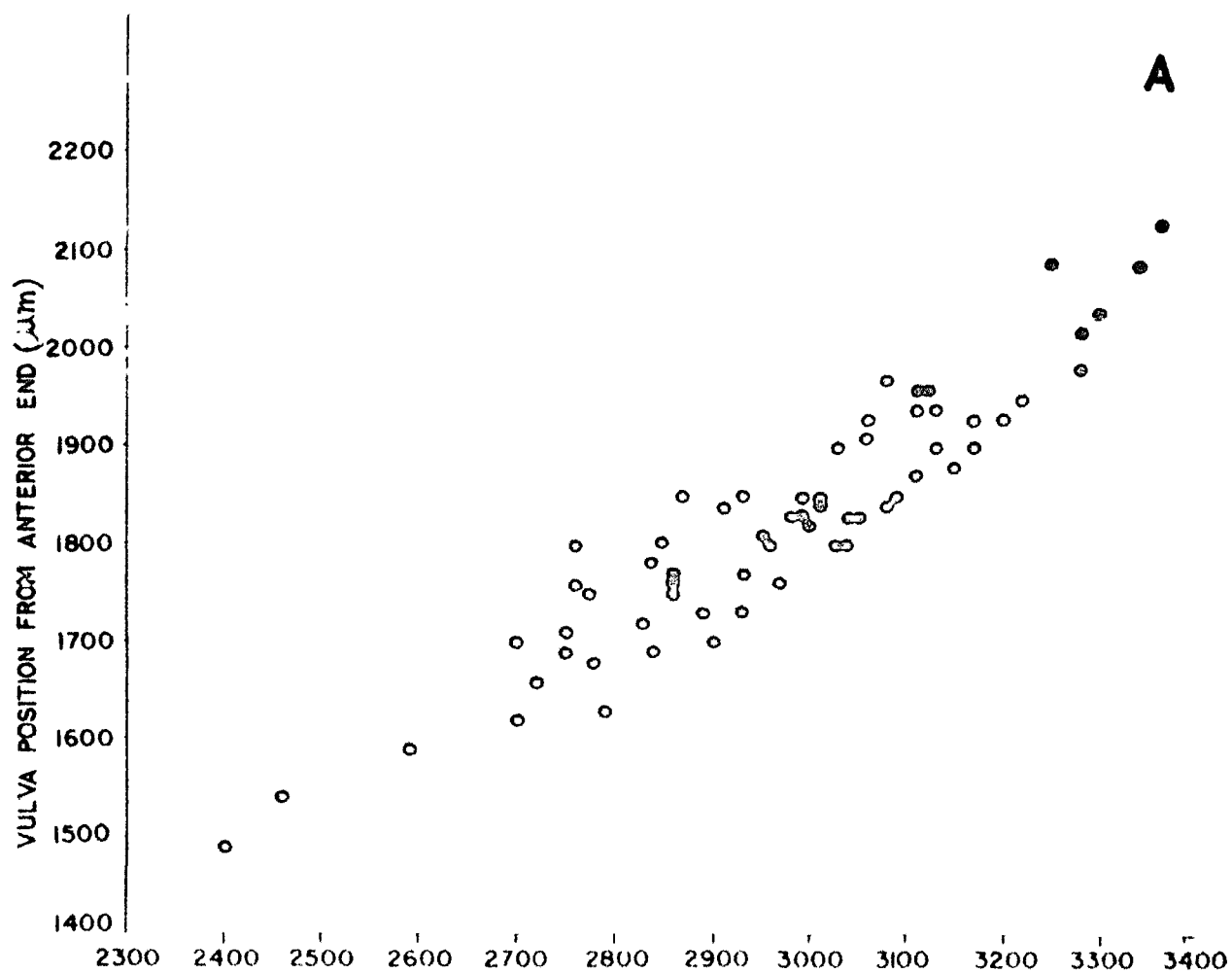


Fig. XIX

The body length in relation to A - buccal cavity length;  
B - oesophageal length; C - tail length in juveniles.

- L<sub>1</sub>
- L<sub>2</sub>
- L<sub>3</sub>
- L<sub>4</sub>



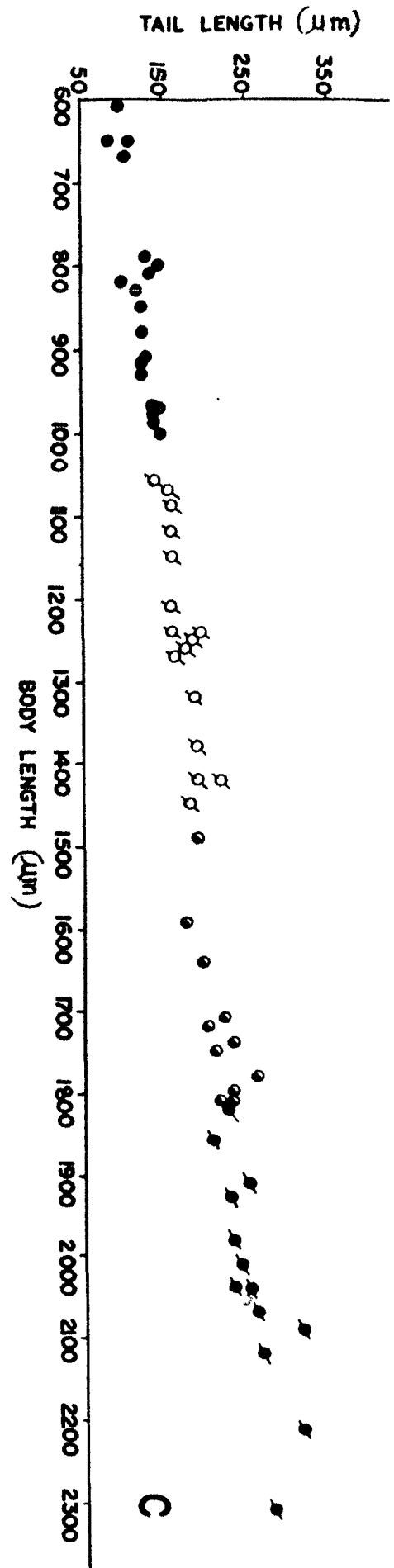
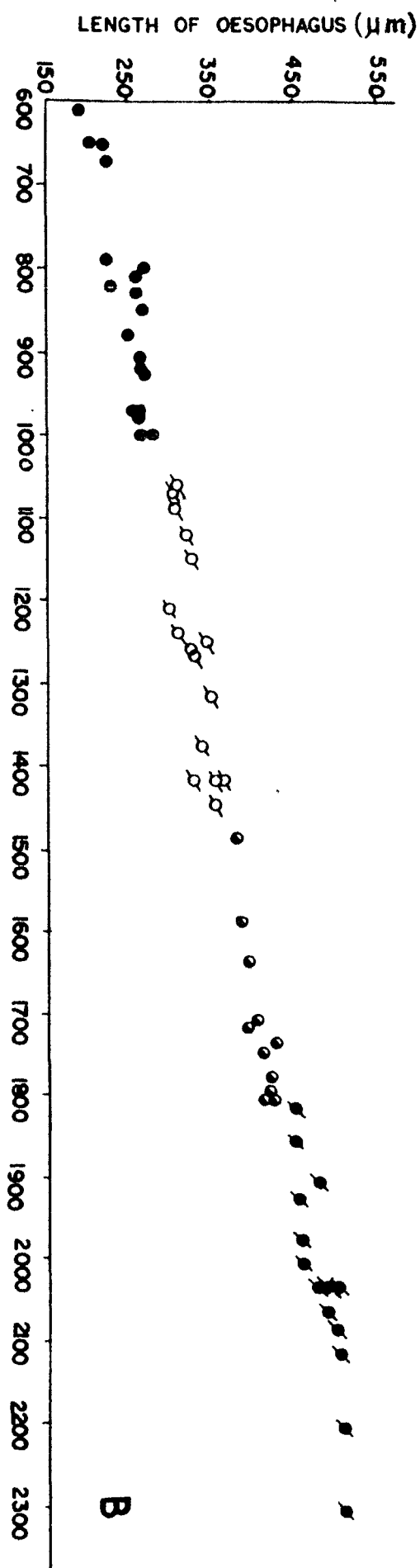
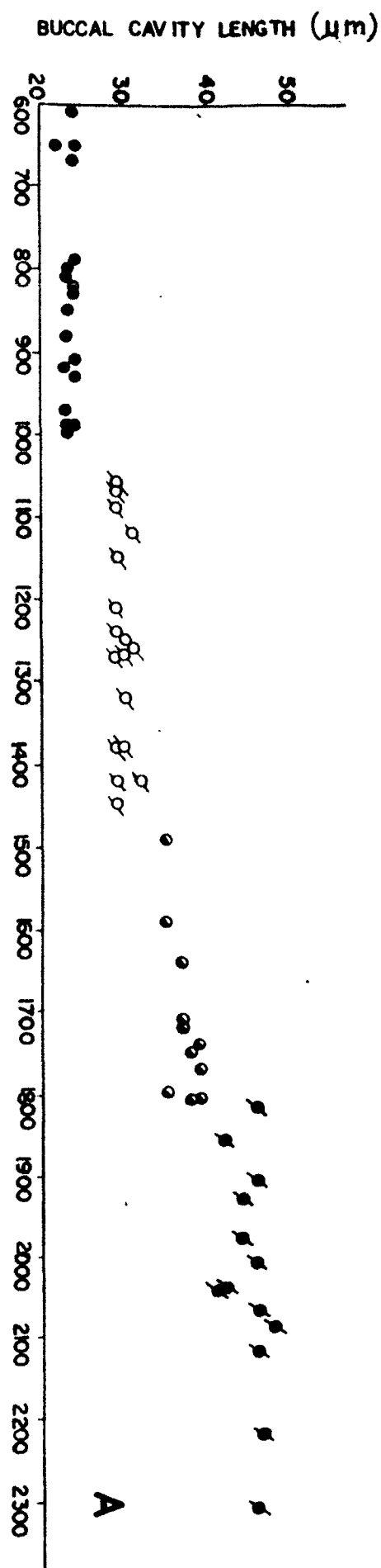
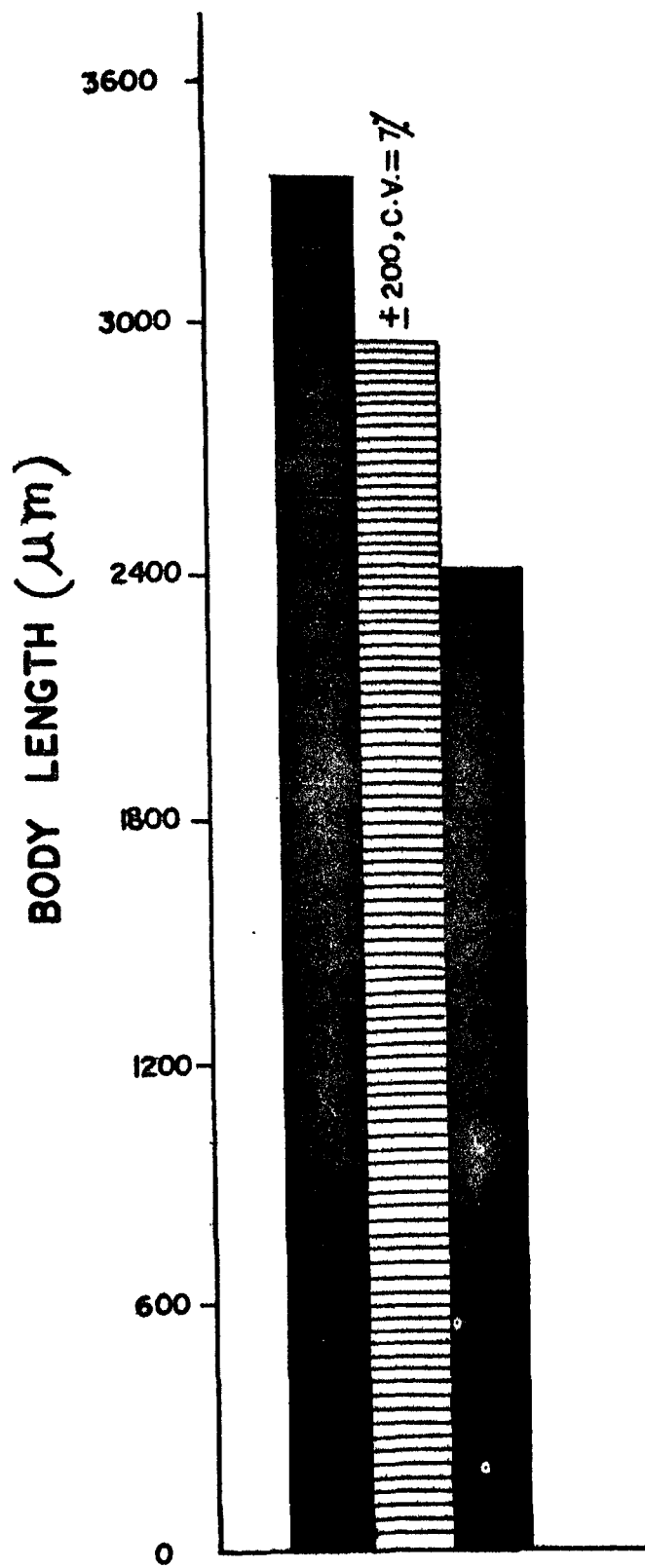
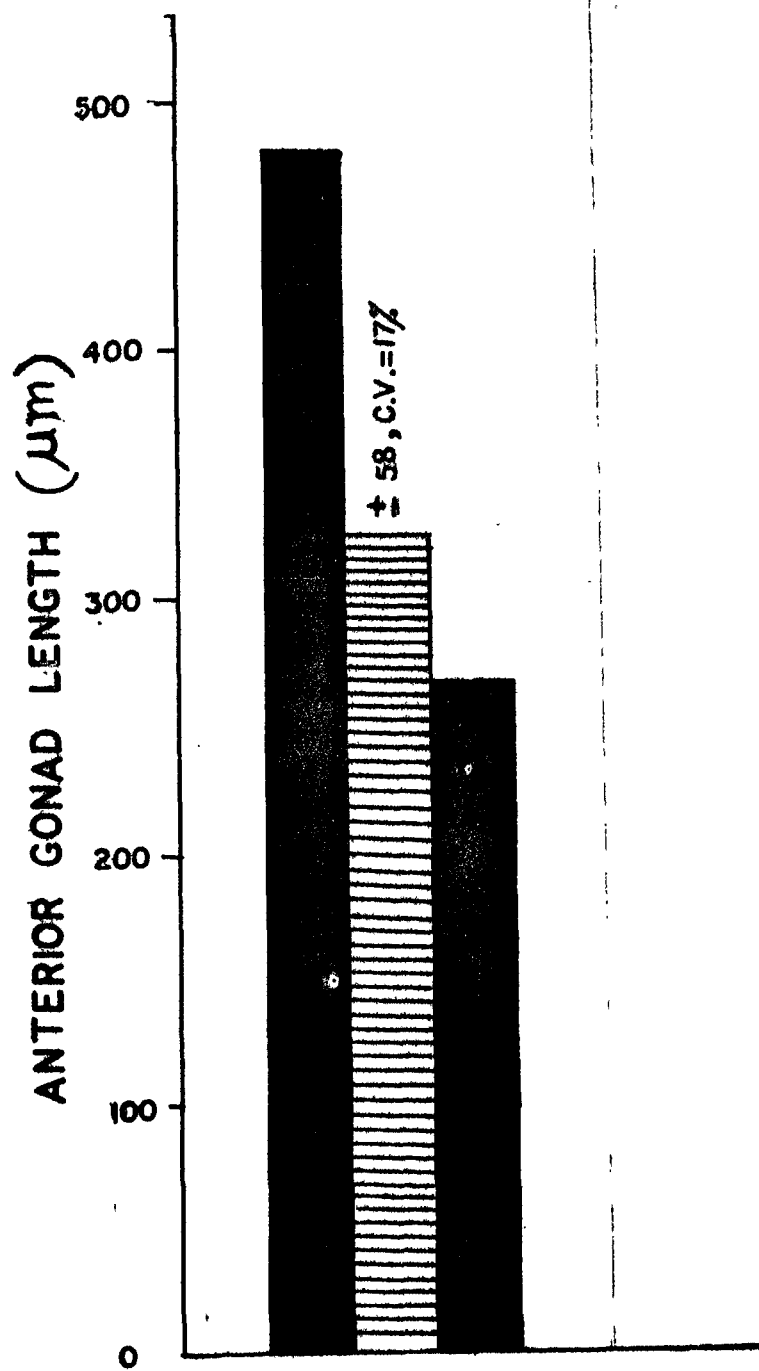


Fig. XX

Histogram showing range, mean, Sd, and CV for A - body length; B - anterior gonad length in adults.



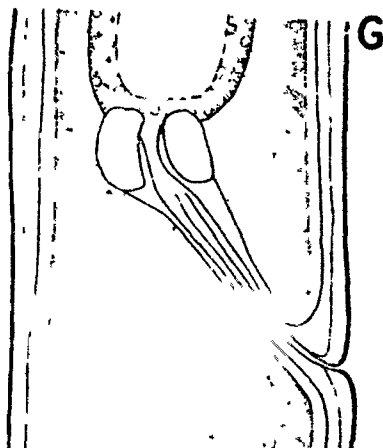
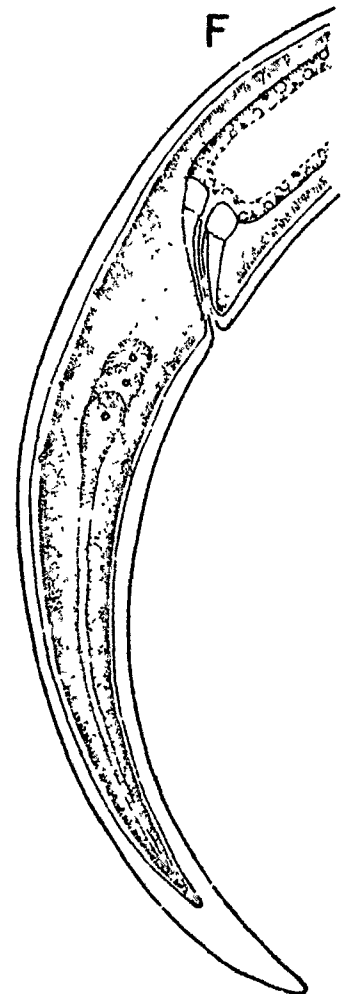
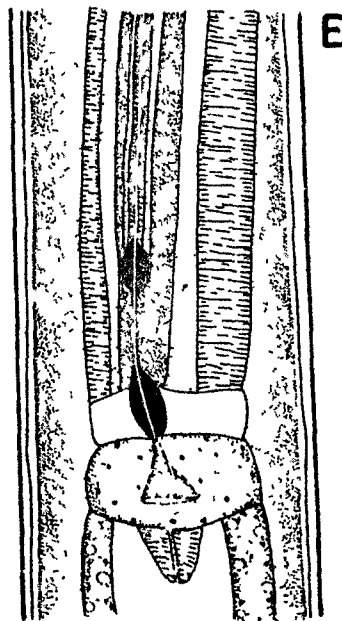
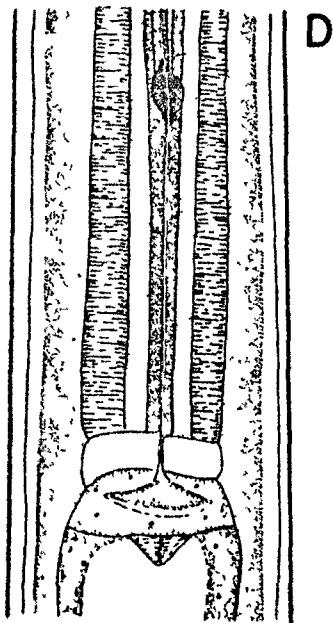
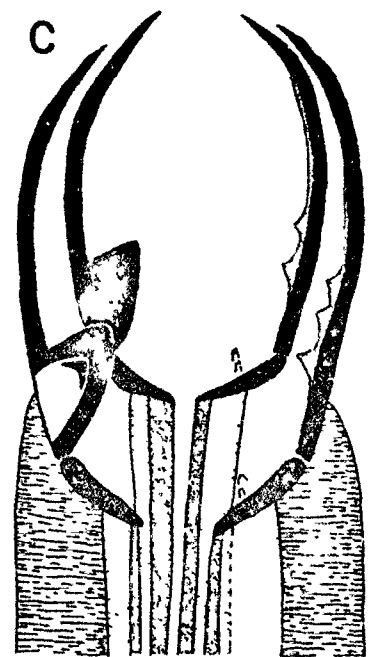
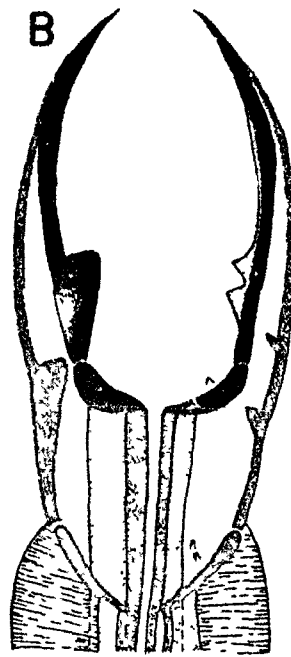
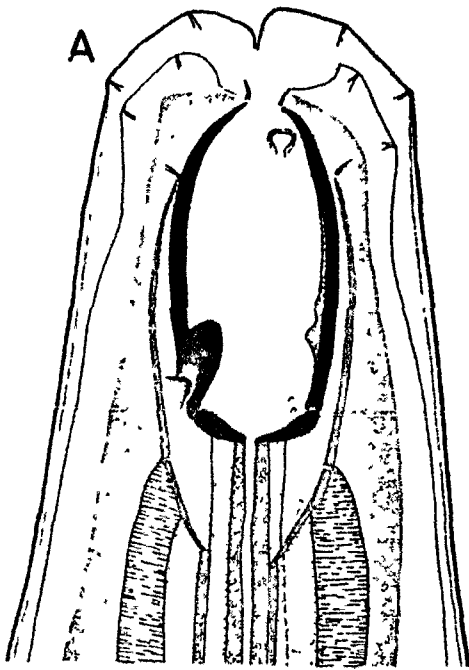
**A**



**B**

Fig. XXI

- A - C - Development of buccal cavity,
- D - Oesophago-intestinal junction without tubercles.  
The older ones are migrating upwards,
- E - Oesophago-intestinal junction with newly formed  
tubercles while the older ones are migrating upwards.
- F - Abnormal tail (third moulting stage),
- G - Anal region (fourth moulting stage).

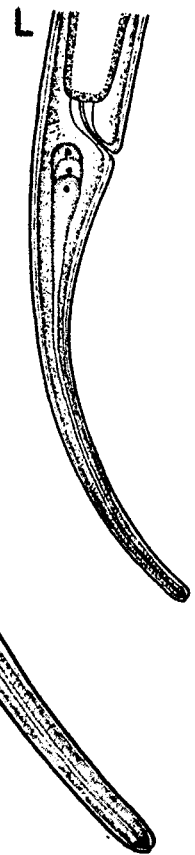
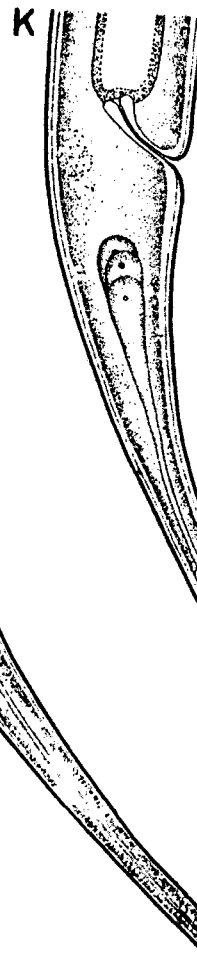
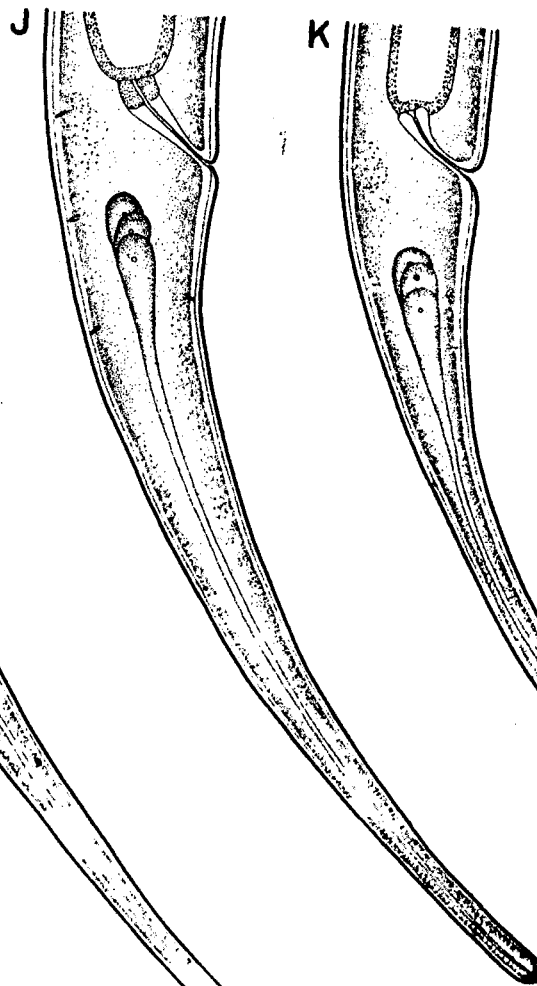
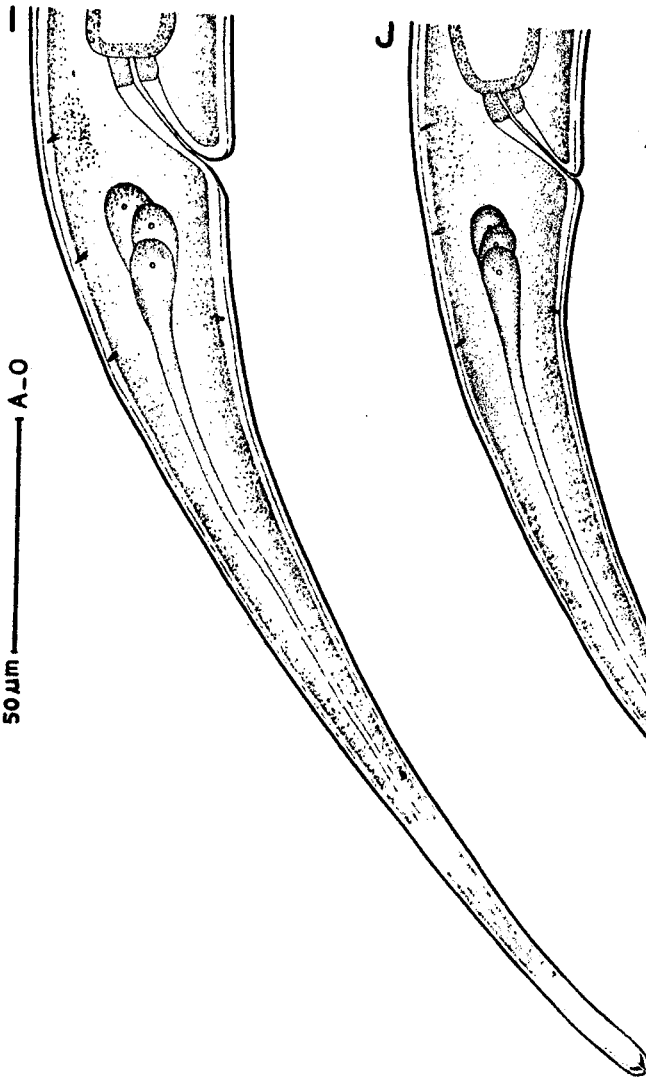
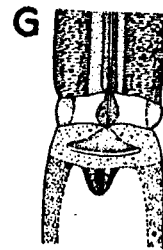
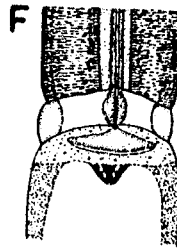
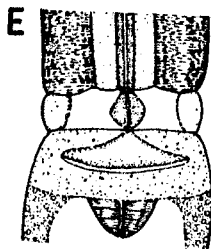
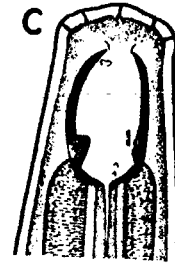
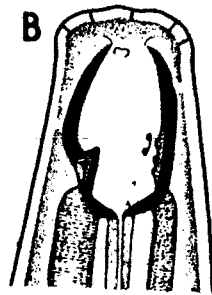
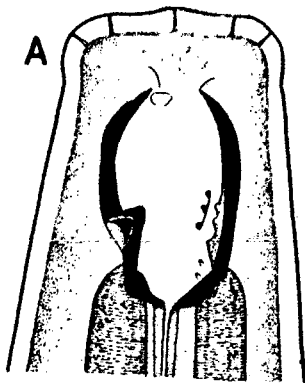


40  $\mu\text{m}$  |—————| D  
 25  $\mu\text{m}$  |—————| A-C, G  
 50  $\mu\text{m}$  |—————| E  
 100  $\mu\text{m}$  |—————| F

Fig. XXII

## JUVENILE STAGES

- A - D - Head ends of  $L_4$ ,  $L_3$ ,  $L_2$ ,  $L_1$  respectively.
- E - H - Oesophago-intestinal junction of  $L_4$ ,  $L_3$ ,  $L_2$ ,  $L_1$  respectively,
- I - L - Tails of  $L_4$ ,  $L_3$ ,  $L_2$ ,  $L_1$  respectively.



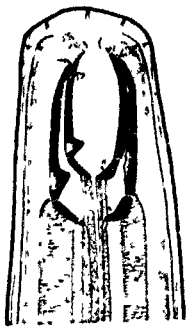
50  $\mu$ m — A-O

Fig. XXIII

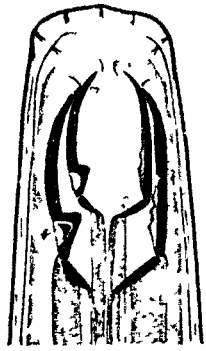
MOULTING STAGES

- A - D - Head ends of first, second, third and fourth moulting stages respectively.
- E - H - Tails of first, second, third and fourth moulting stages respectively.

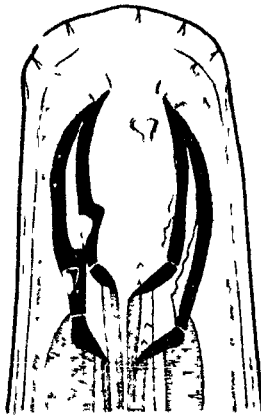




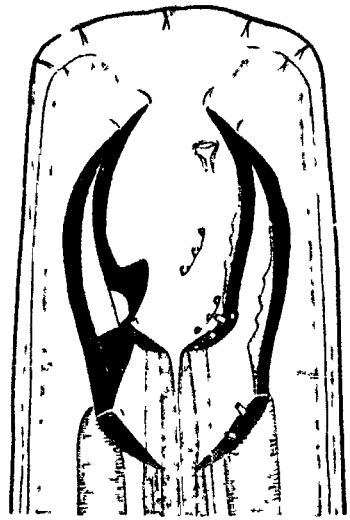
A



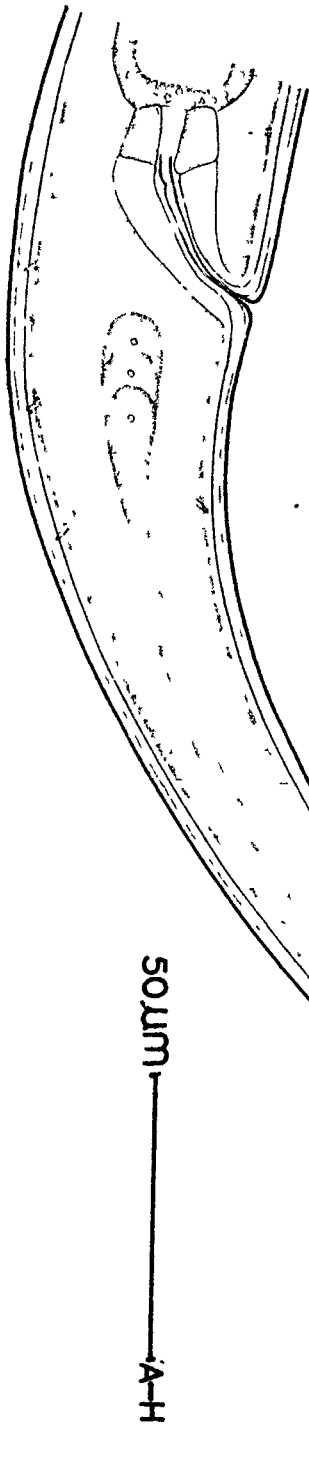
B



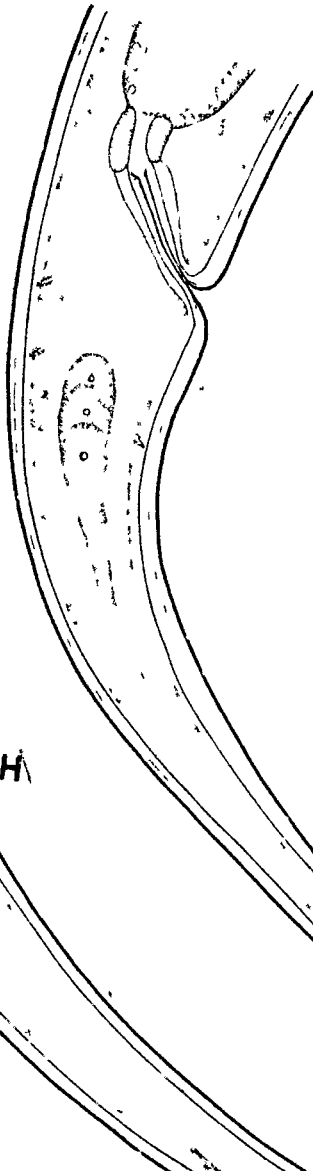
C



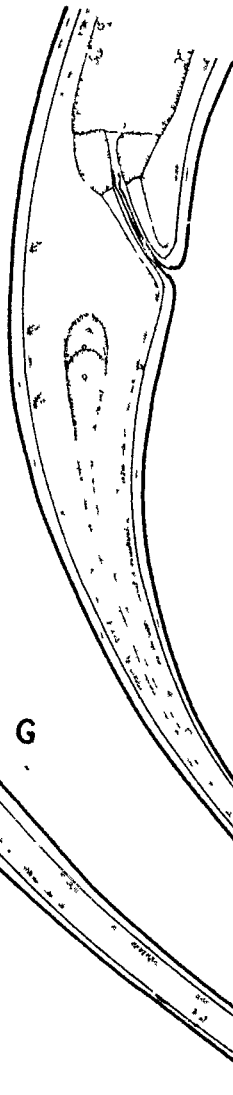
D



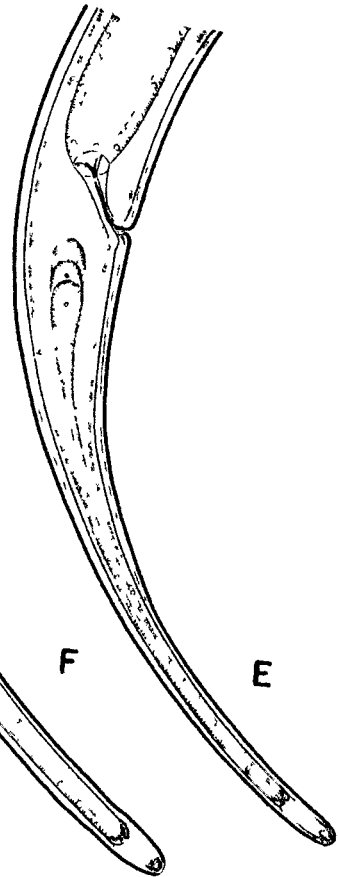
H



G



F



E

50µm  
A-H

Fig. XXIV

## DEVELOPMENT OF THE FEMALE GONADS

- |   |   |                              |
|---|---|------------------------------|
| A | - | First stage juvenile,        |
| B | - | Second stage juvenile,       |
| C | - | Second moulting stage,       |
| D | - | Third stage juvenile,        |
| E | - | Third moulting stage,        |
| F | - | Fourth stage juvenile,       |
| G | - | Early fourth moulting stage, |
| H | - | Late fourth moulting stage.  |

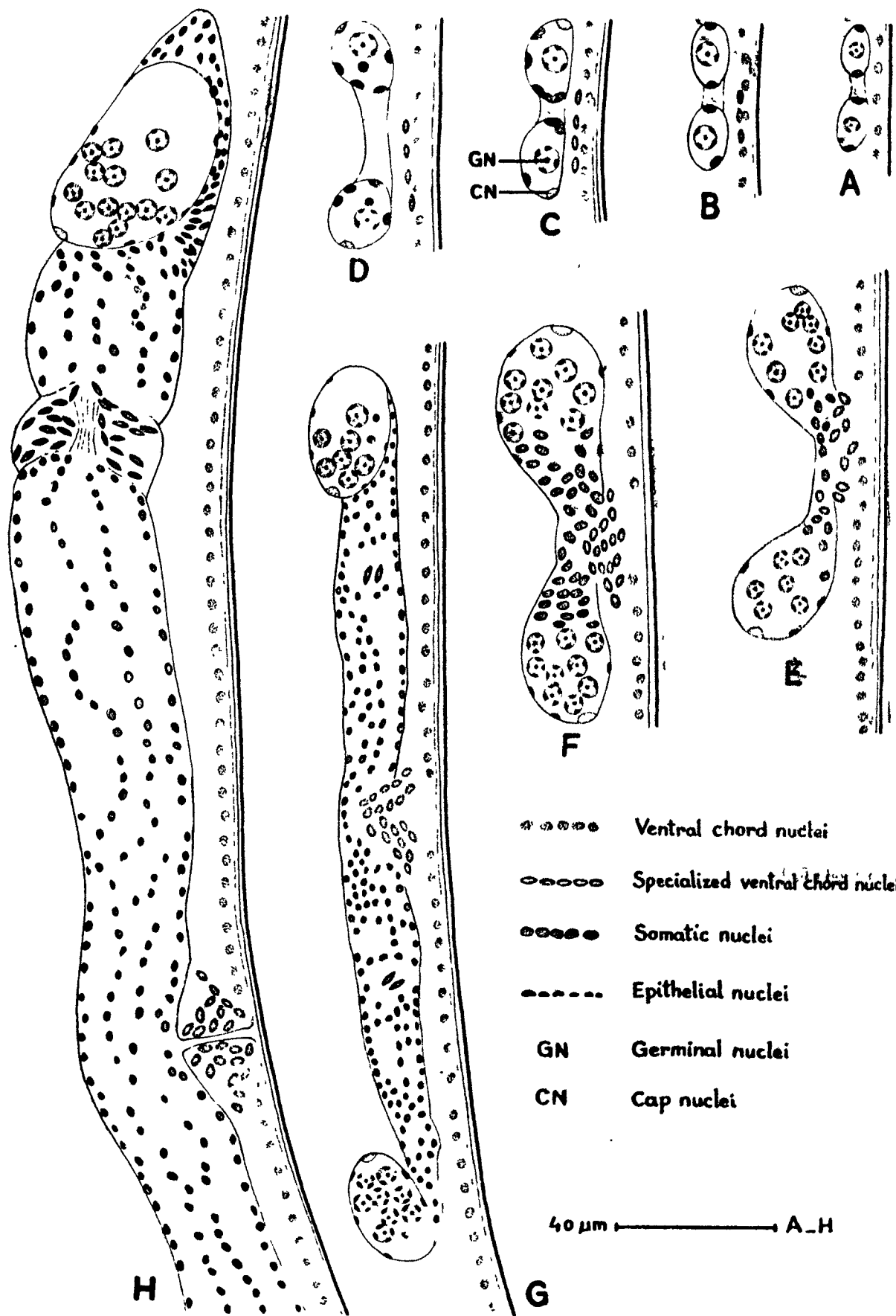


Fig. XXV

## DEVELOPMENT OF THE MALE GONADS

- |       |   |                         |
|-------|---|-------------------------|
| A     | - | Second stage juveniles, |
| B     | - | Second moulting stage,  |
| C     | - | Third stage juvenile,   |
| D     | - | Third moulting stage,   |
| E & F | - | Fourth stage juvenile,  |
| G     | - | Fourth moulting stage,  |
| H     | - | Third stage male tail,  |
| I     | - | Fourth stage male tail. |

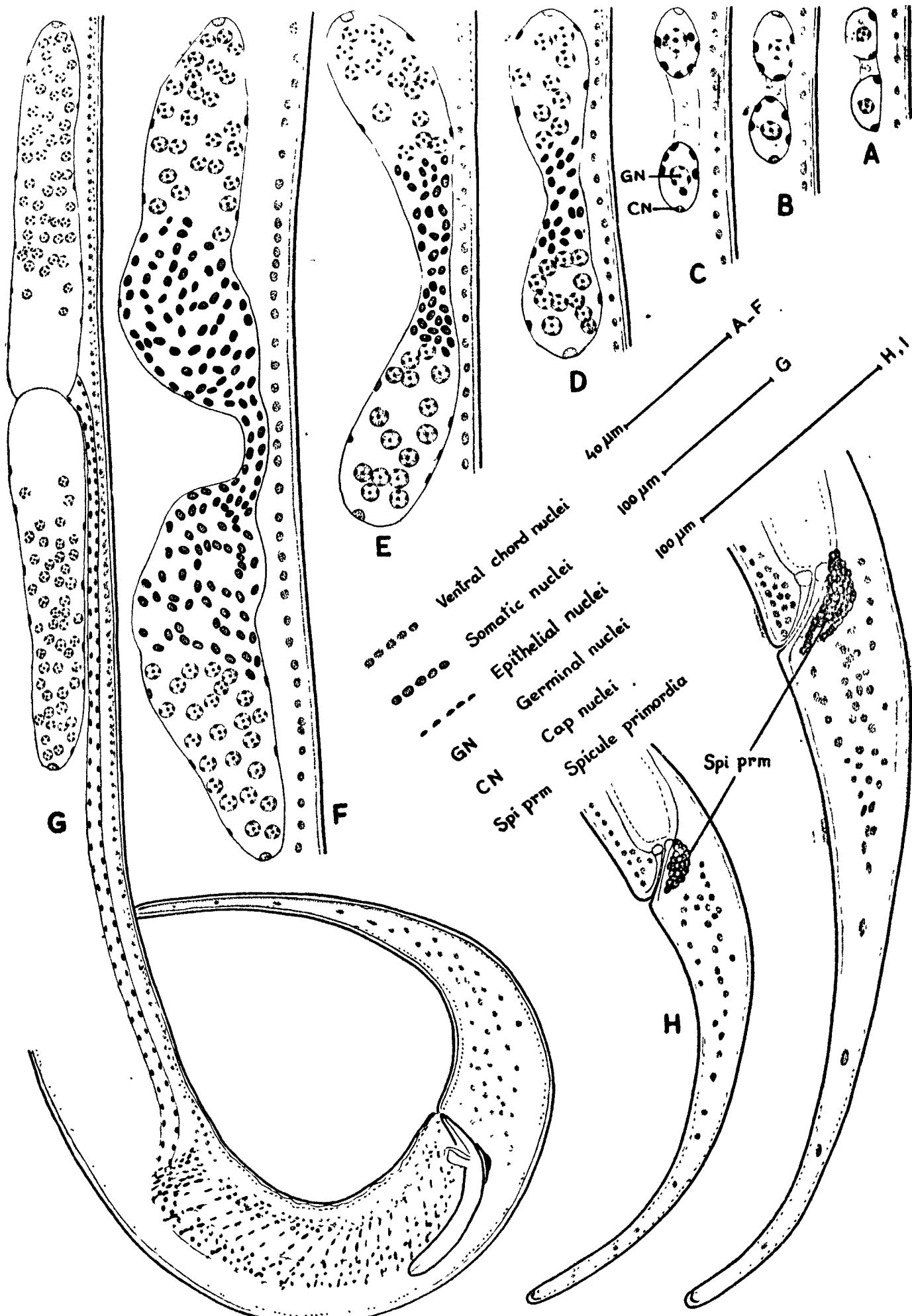


Fig. XXVI

- A - Length of ovary in different months,
- B - Length of genital tract in different months.

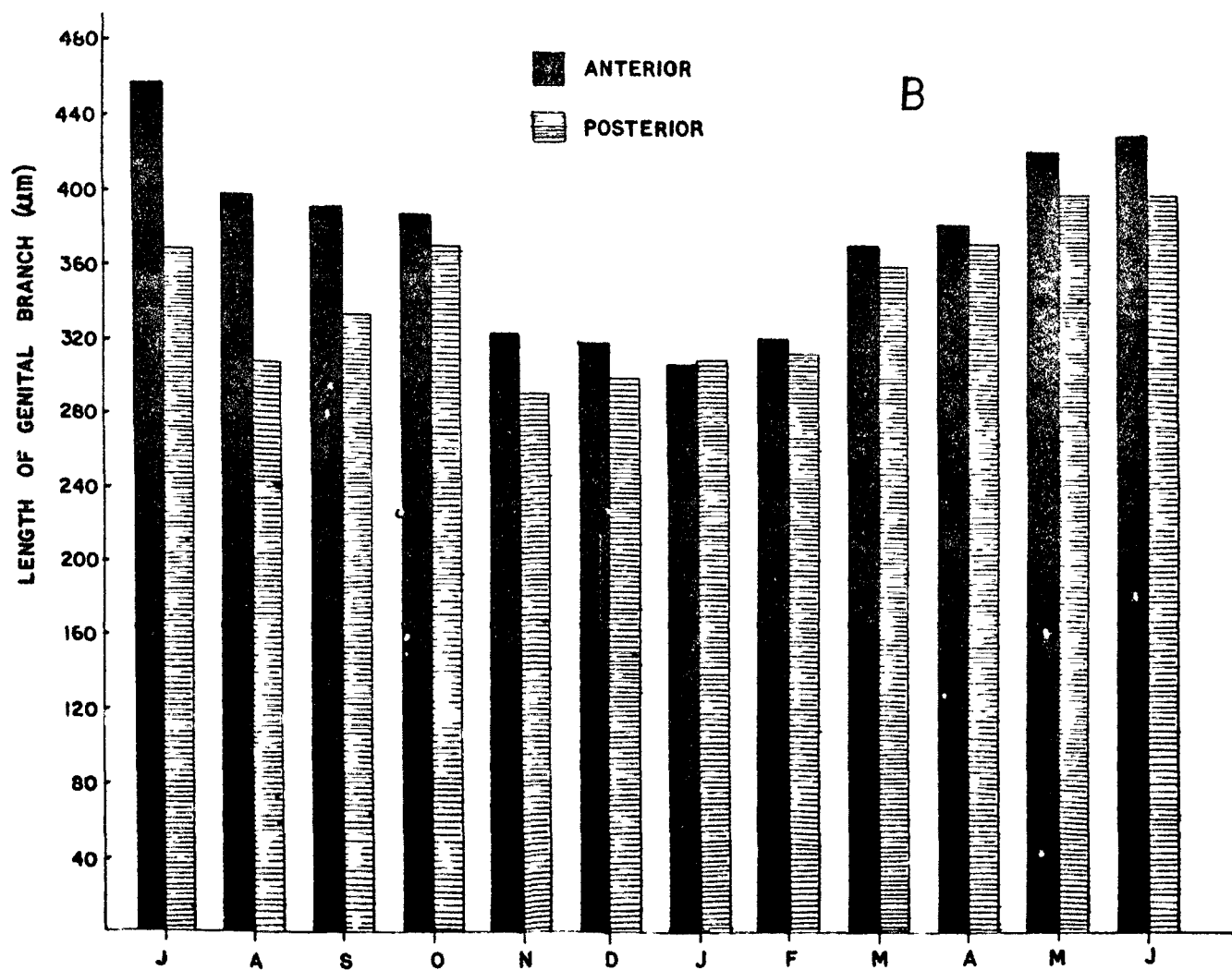
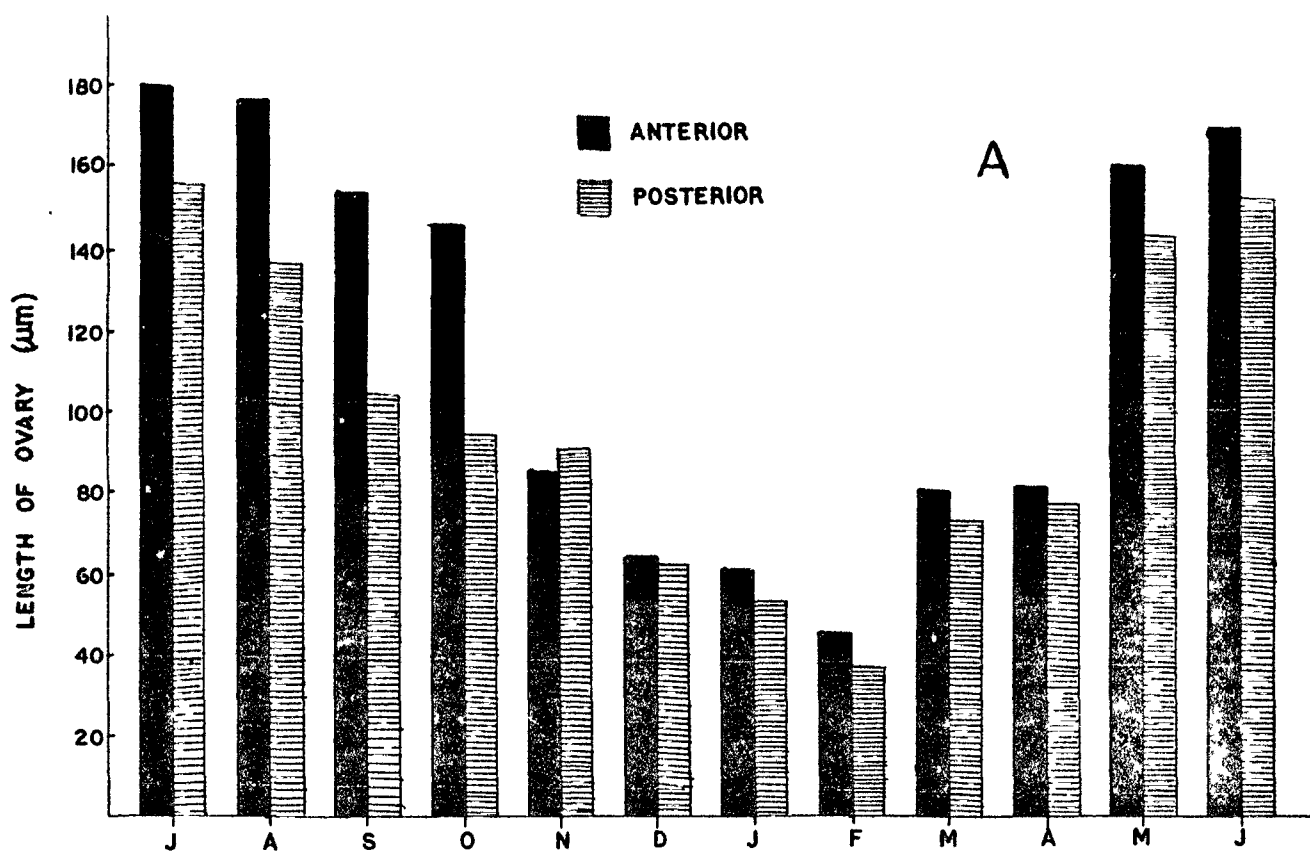


Fig. XXVII

Seasonal fluctuation in A - Soil temperature; B - Soil moisture at Company Garden, Bareilly.

Fig. XXVIII

Seasonal fluctuations in the distribution of Hadronchus shakili in soil.



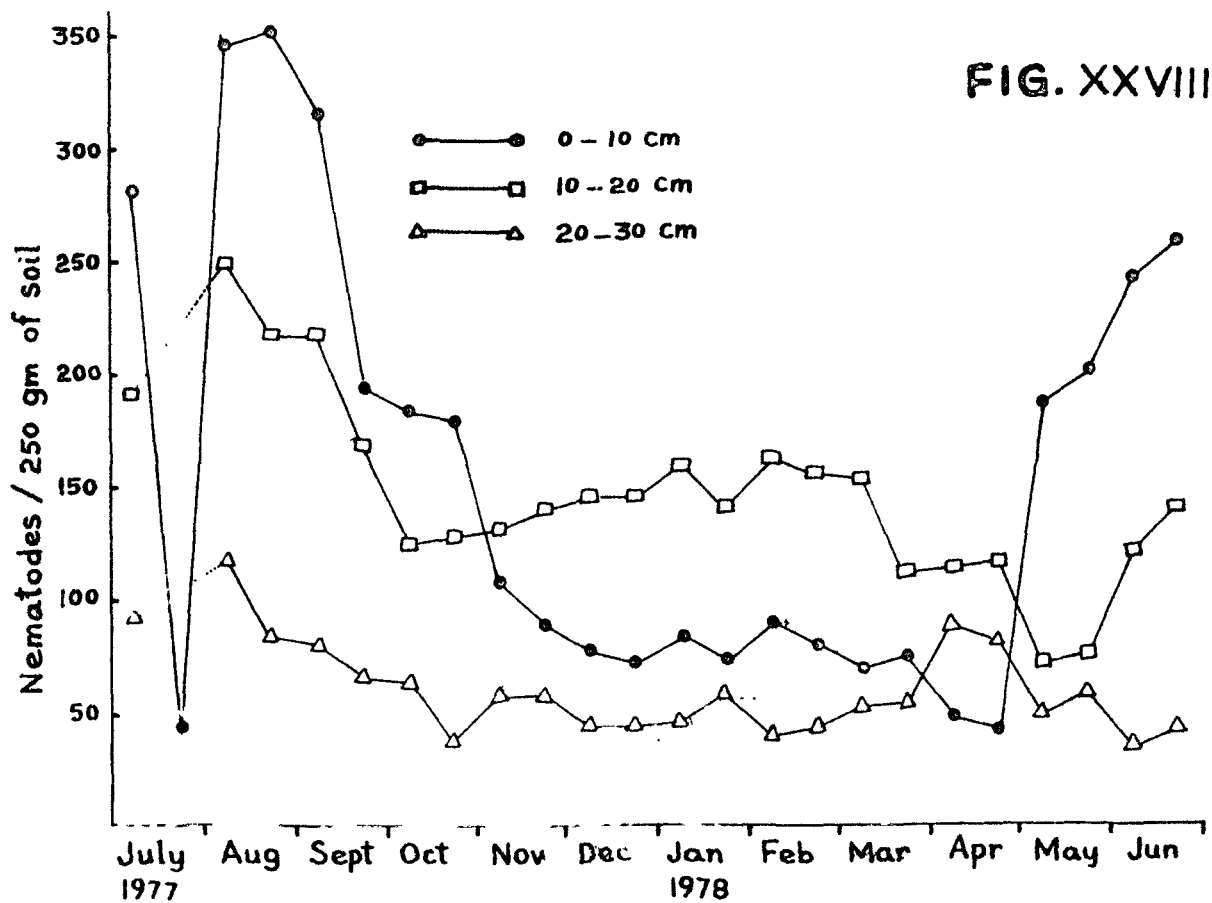
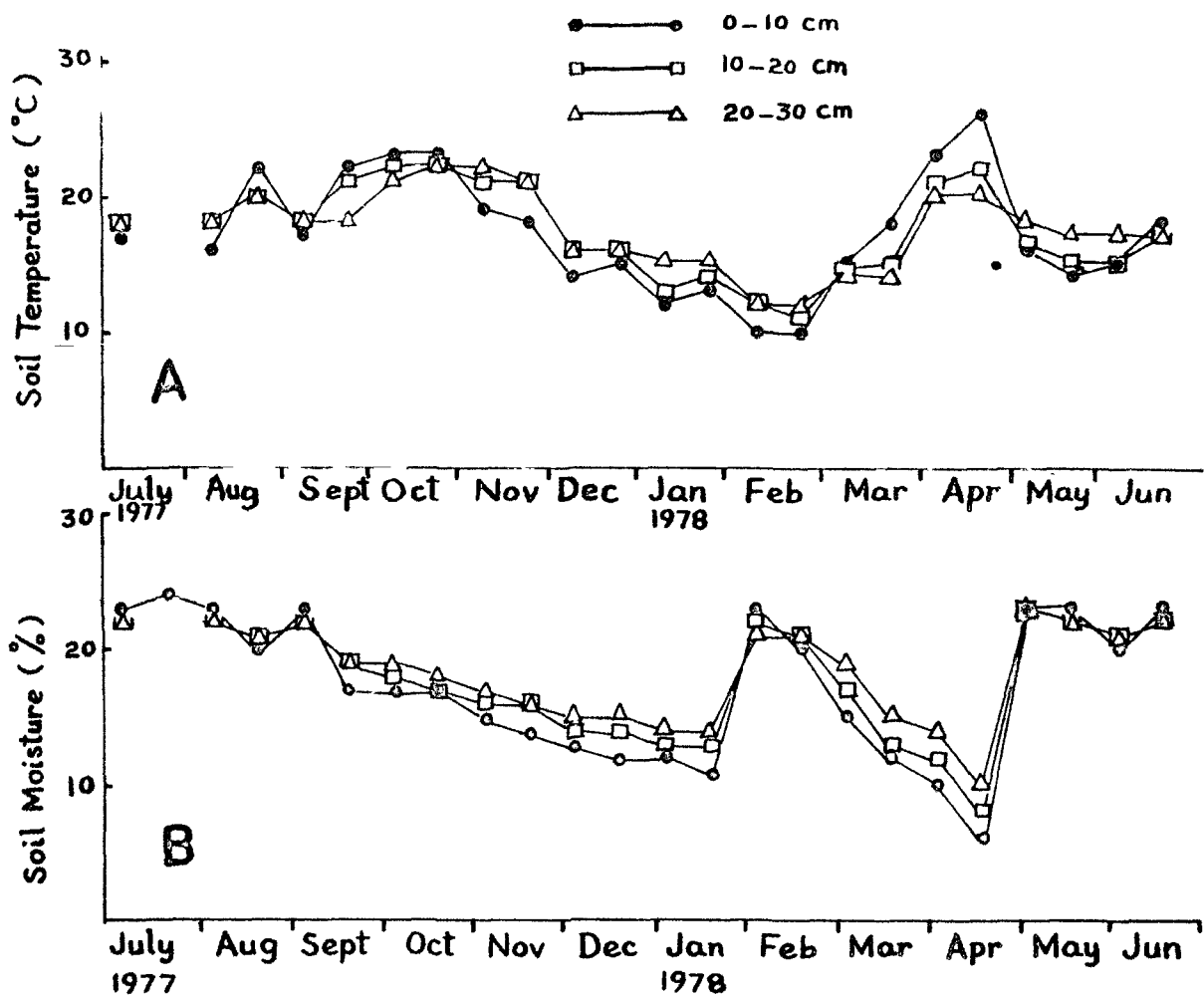


Fig. XXIX

Seasonal fluctuations in the distribution of  $L_1$ ,  $L_2$ ,  $L_3$   
in soil. A - 0-10 cm; B - 10-20 cm; C - 20-30 cm.

FIG. XXIX

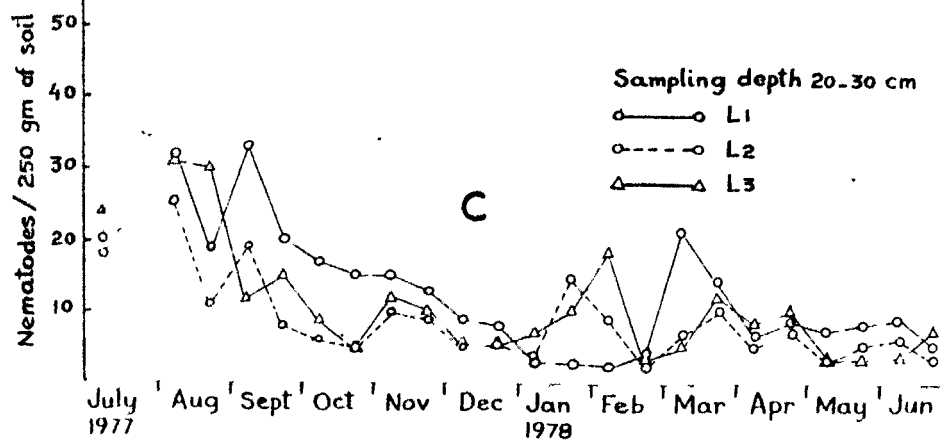
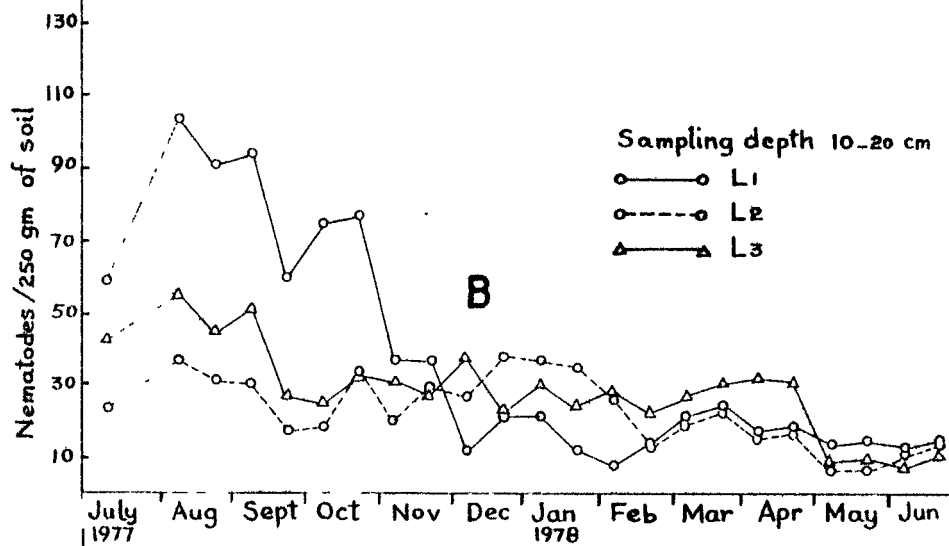
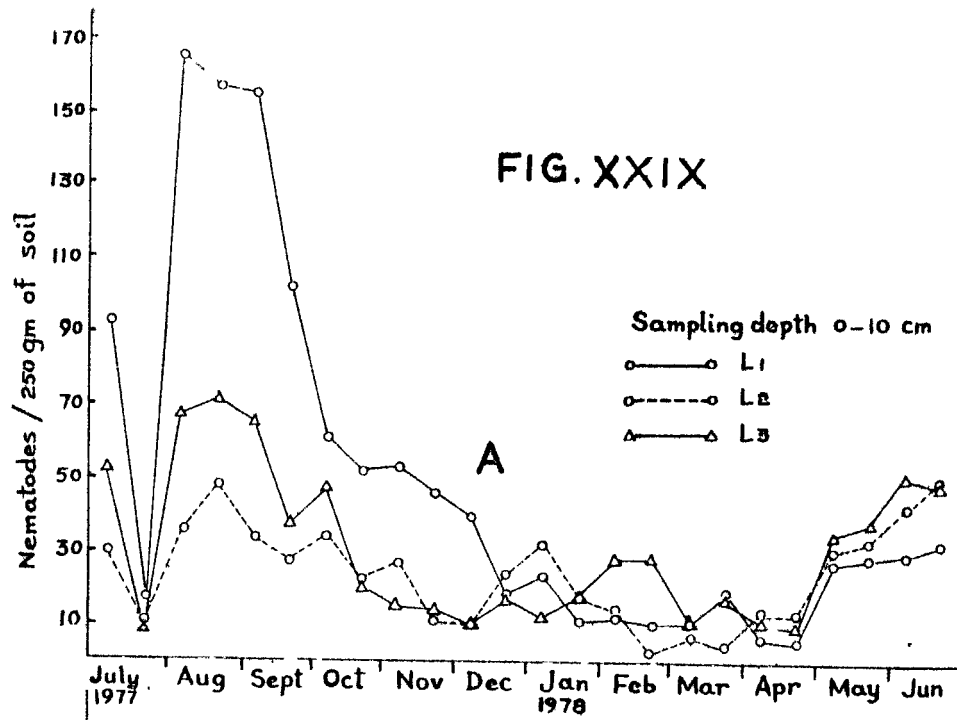


Fig. XXX

Seasonal fluctuations in the distribution of  $L_4$  and adults. A - 0-10 cm; B - 10-20 cm; C - 20-30 cm.

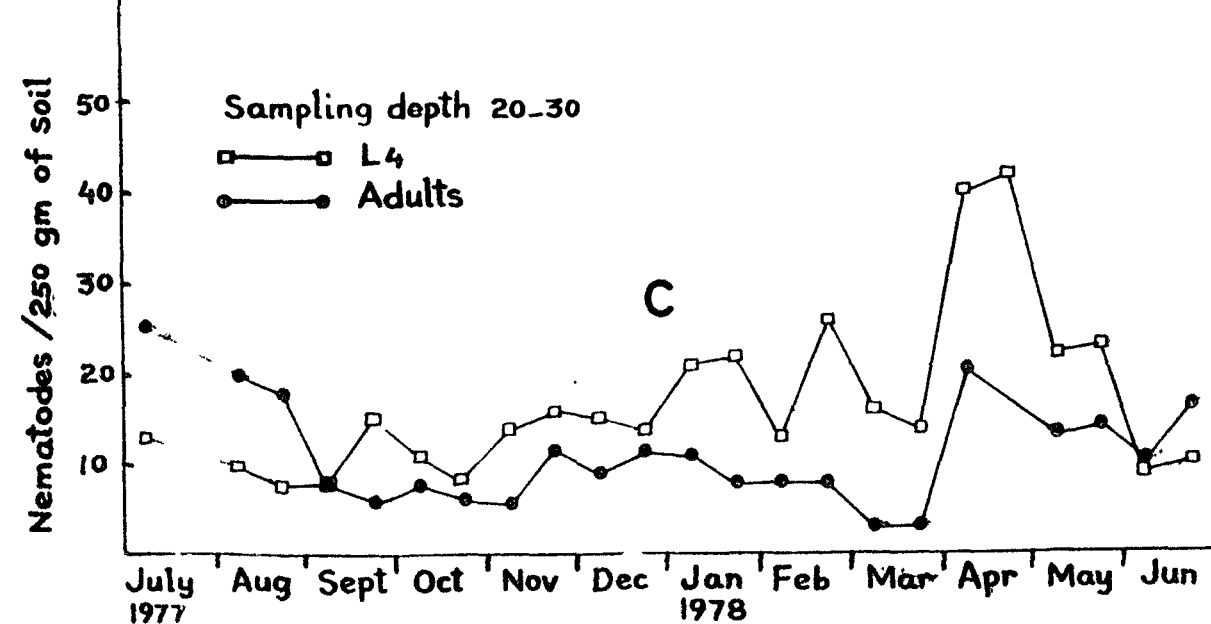
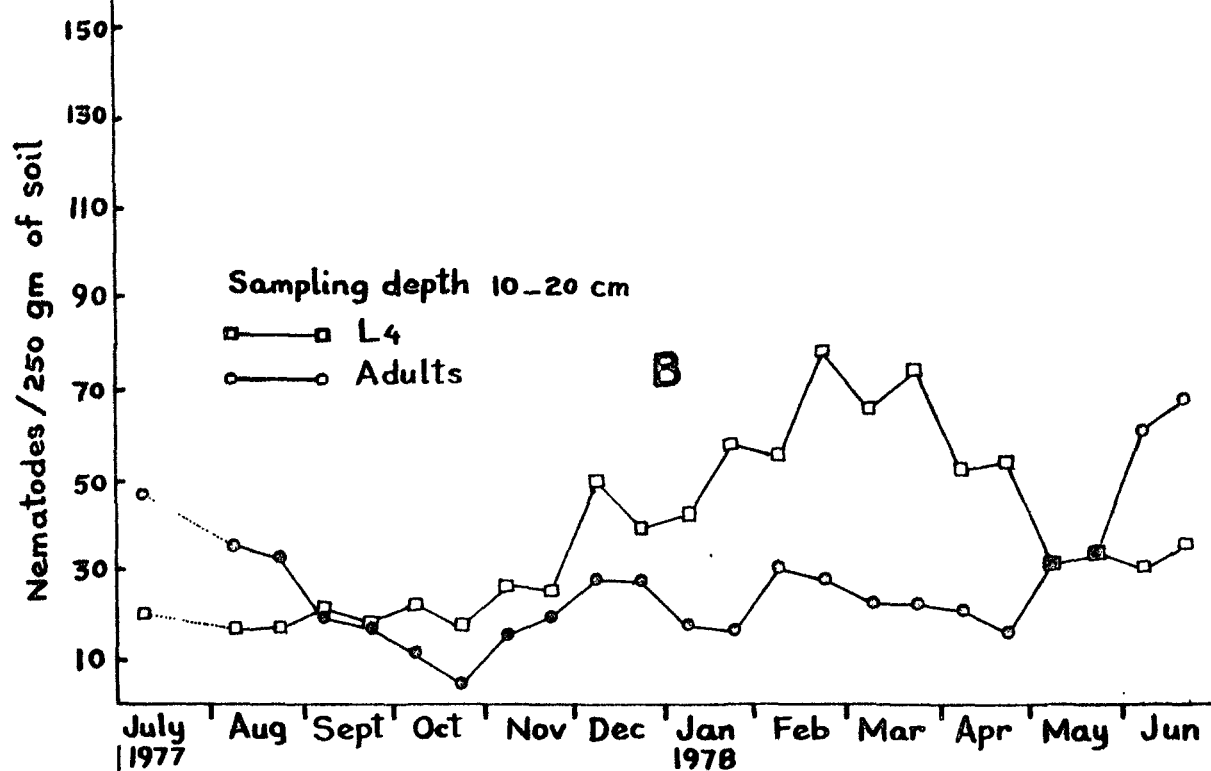
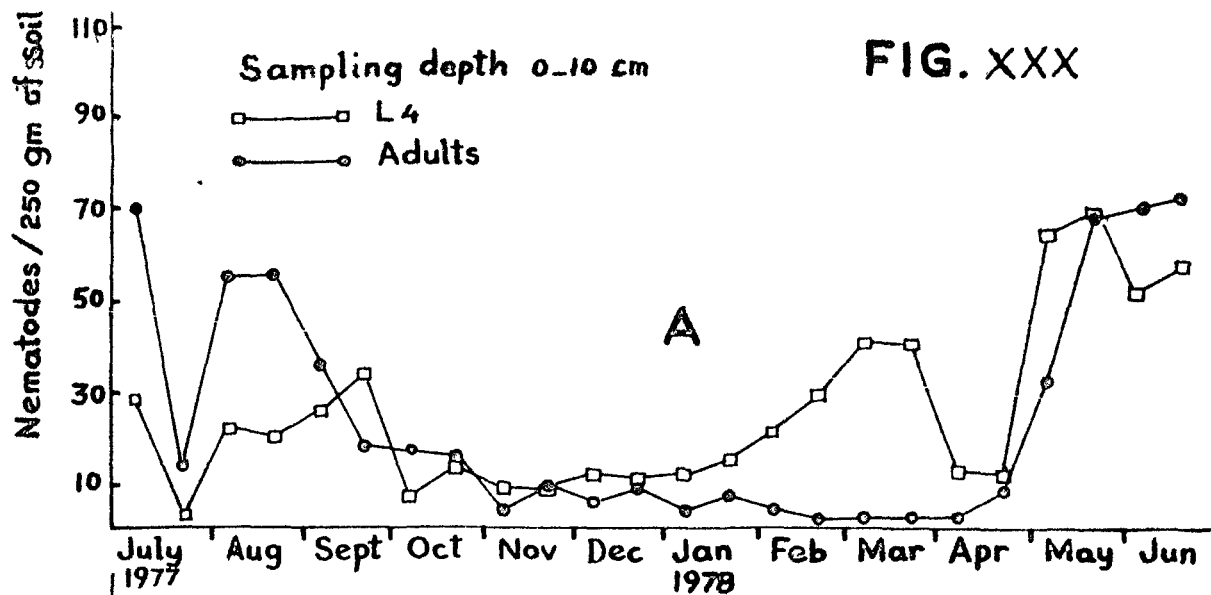
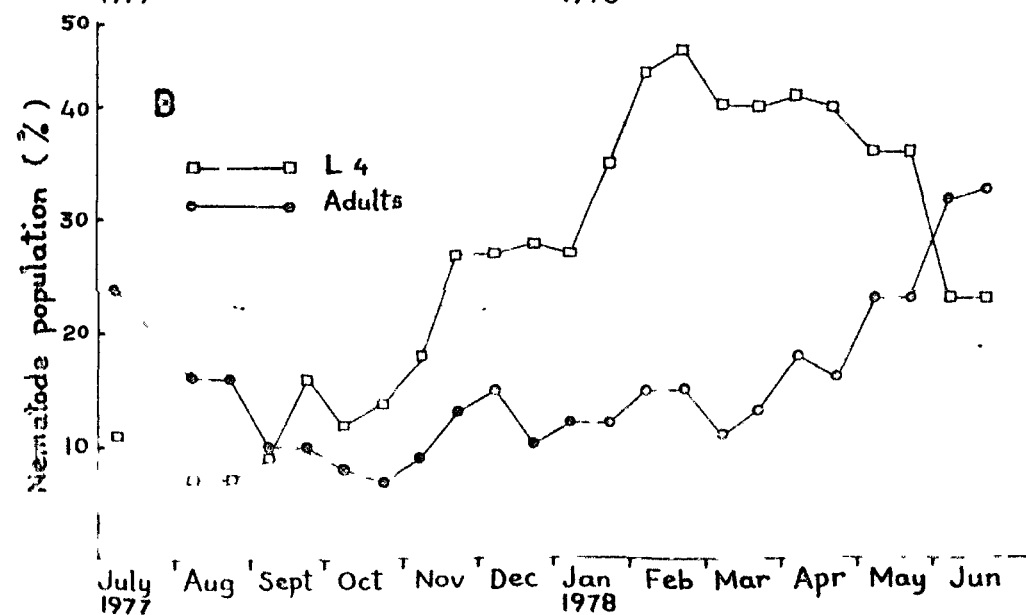
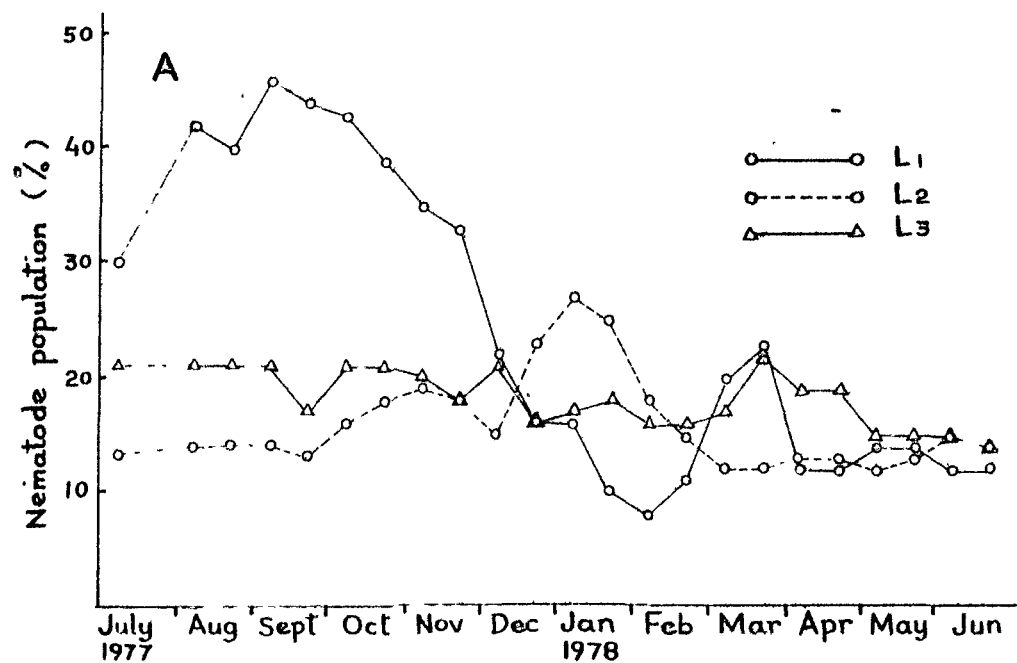
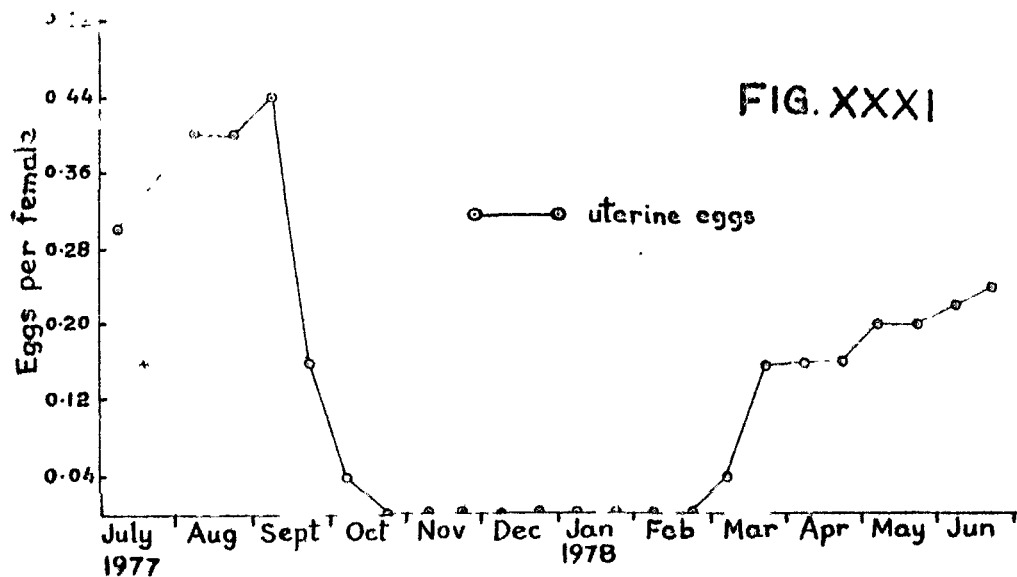


Fig. XXXI

Seasonal fluctuation in uterine egg count.

Fig. XXXII

Seasonal fluctuations in the population of A -  $L_1$ ,  $L_2$ ,  $L_3$ ; B -  $L_4$ , adults.



**FIG. XXXII**

Fig. XXXIII

Relationship between population of A - Hadronchus shakili and the populations of Trichodorus sp. Thornenema sp. and Hemicriconemoides sp., B - H. shakili and the total population of prey (other nematodes).



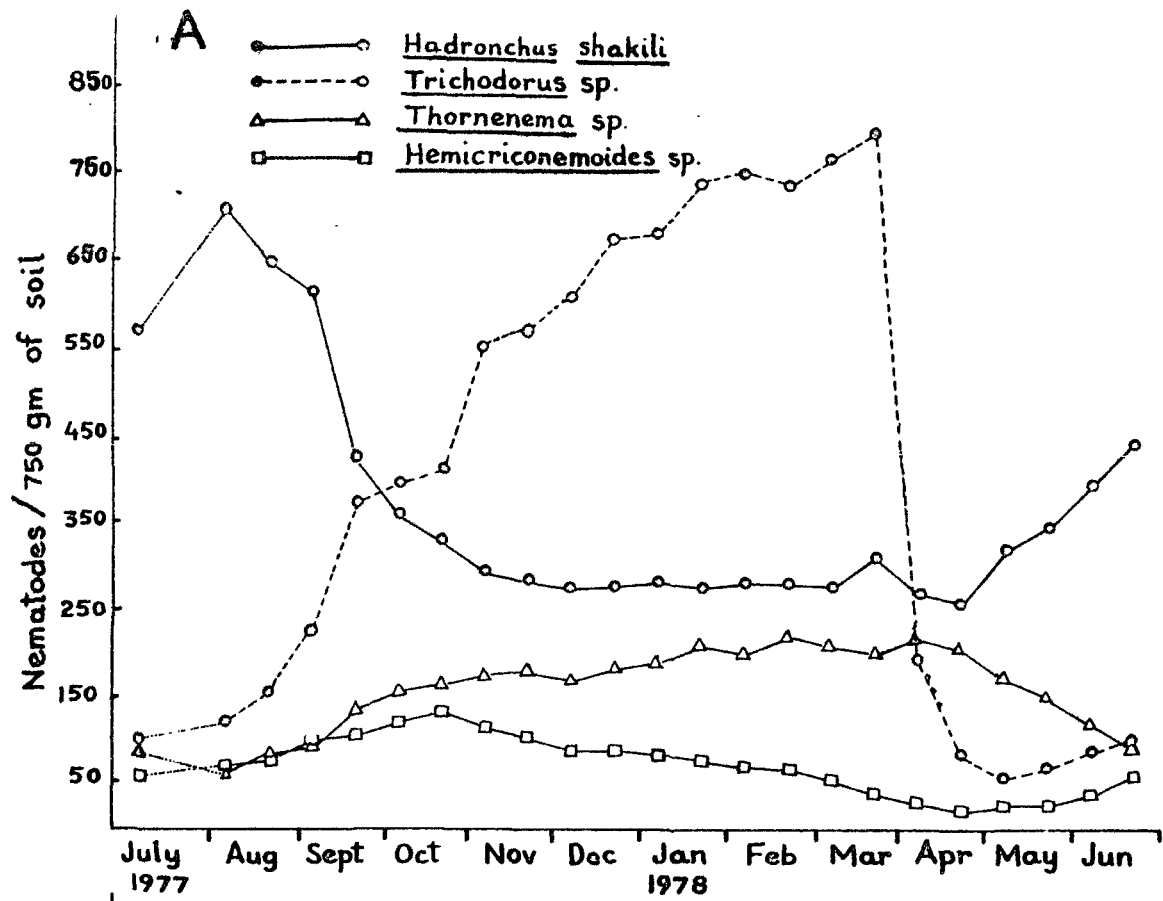


FIG. XXXIII

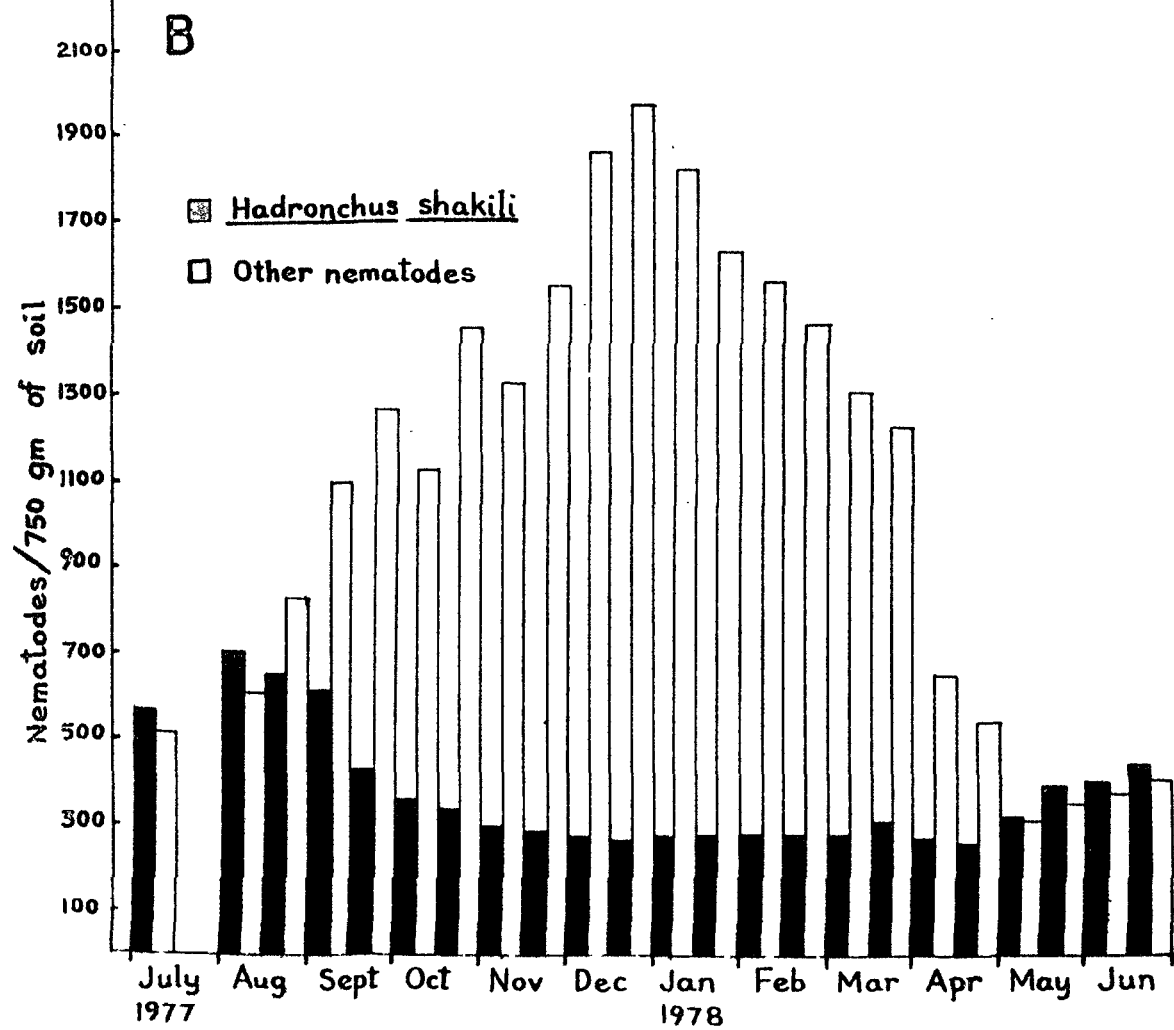


Fig. XXXIV

Survival time of adults and juveniles in A - tap water;  
B - phosphate buffer of different pH.

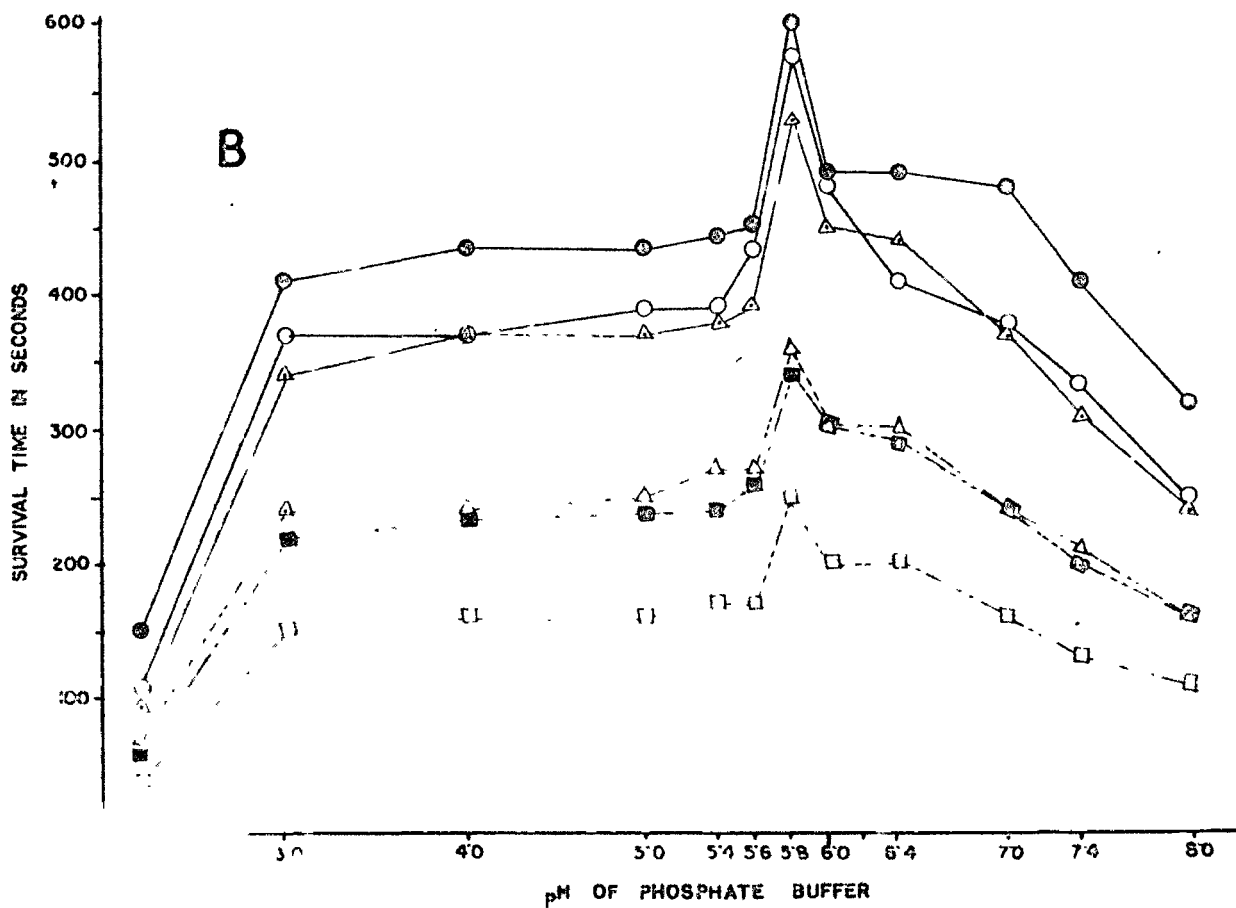
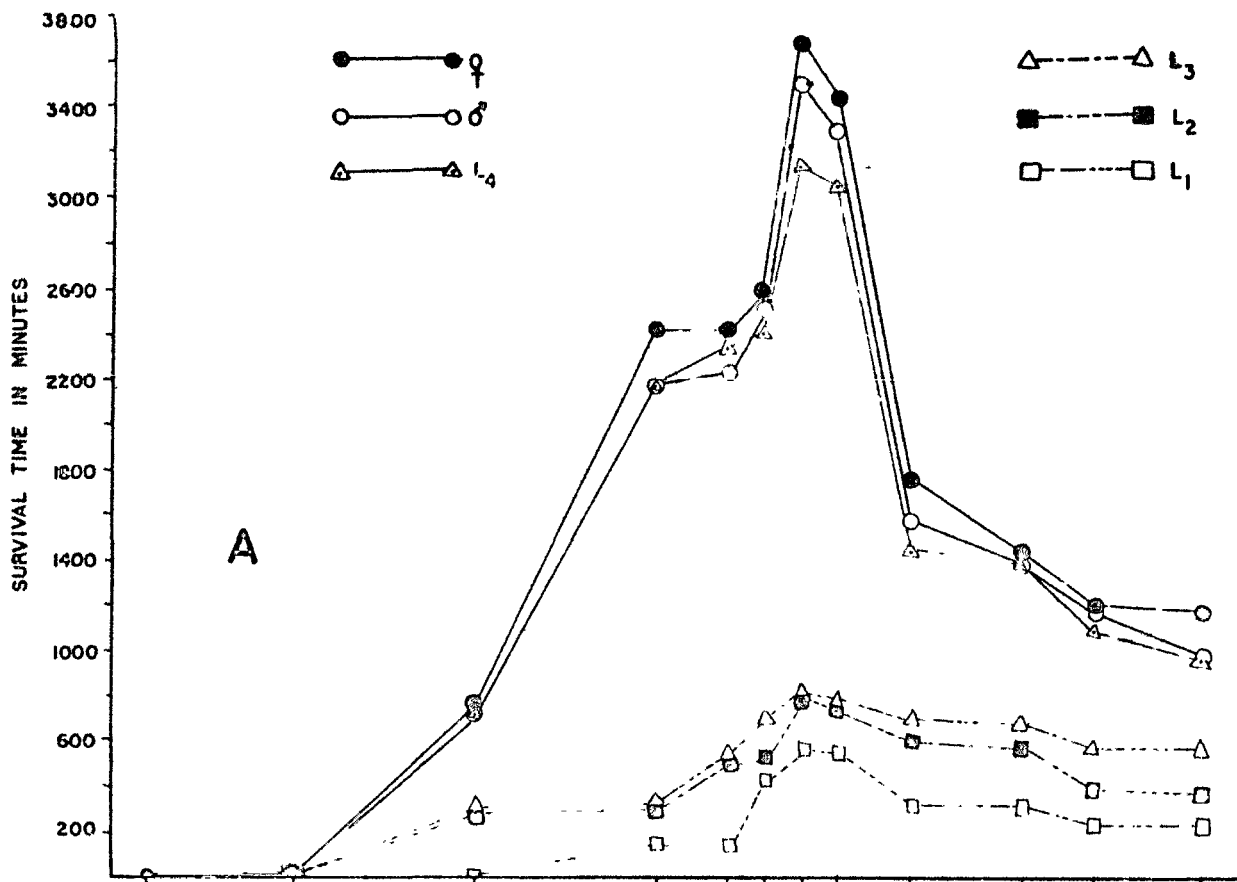


Fig. XXXV

Survival time of adults and juveniles at different concentrations of salts. A - Potassium carbonate; B - Potassium chloride; C - Potassium nitrate; D - Copper sulphate.

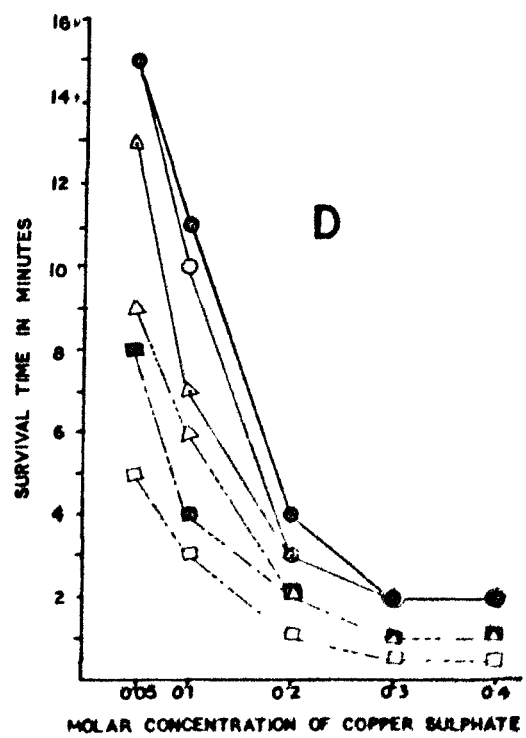
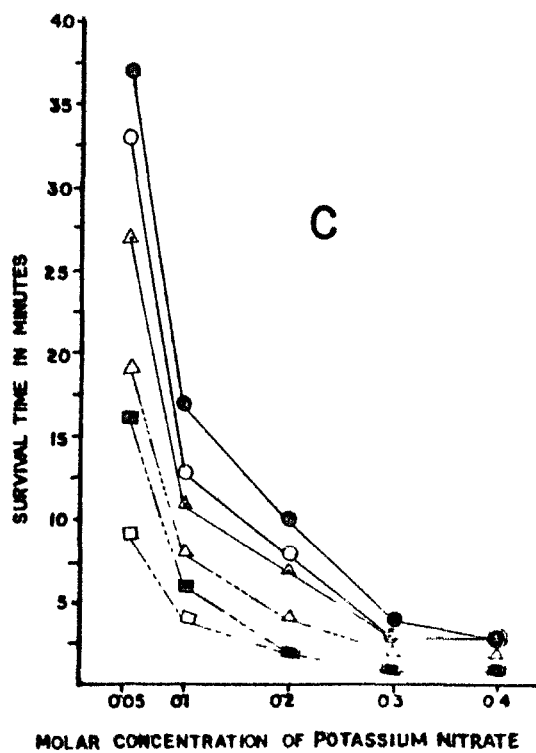
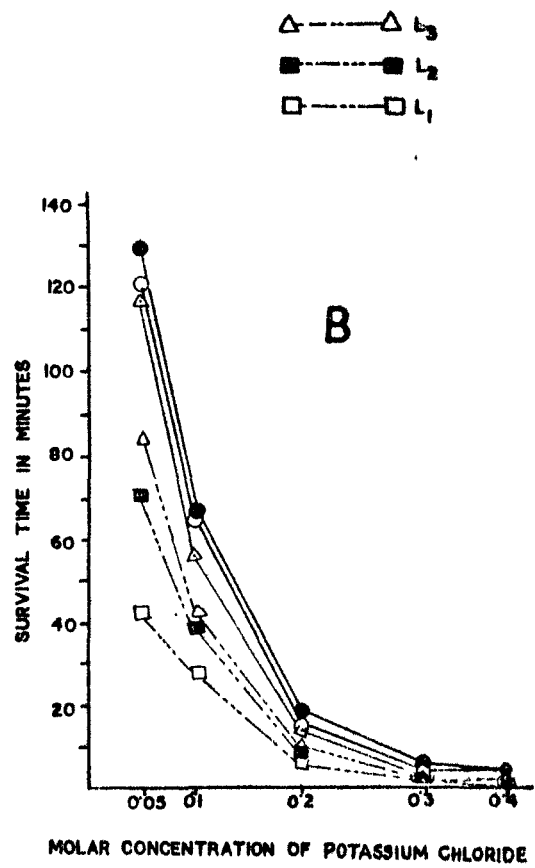
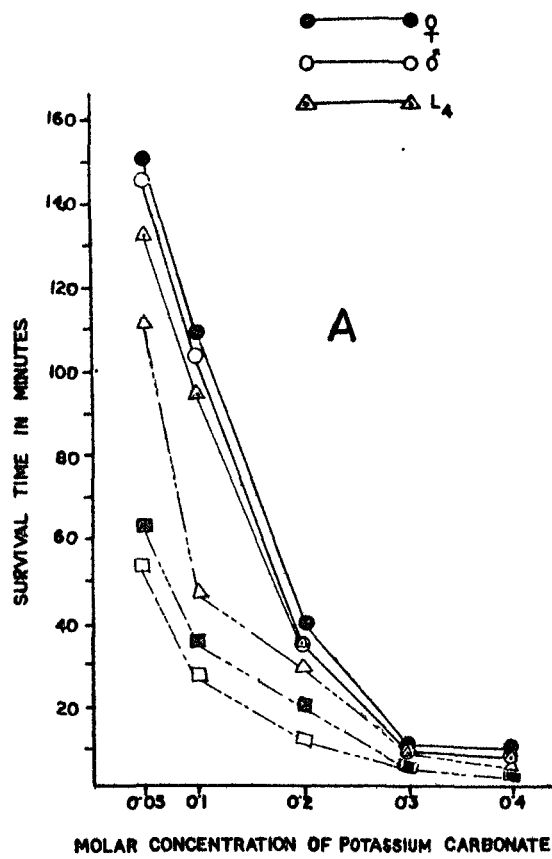


Fig. XXXVI

Survival time of adults and juveniles at different concentrations of acid. A - Acetic acid; B - Propionic acid.

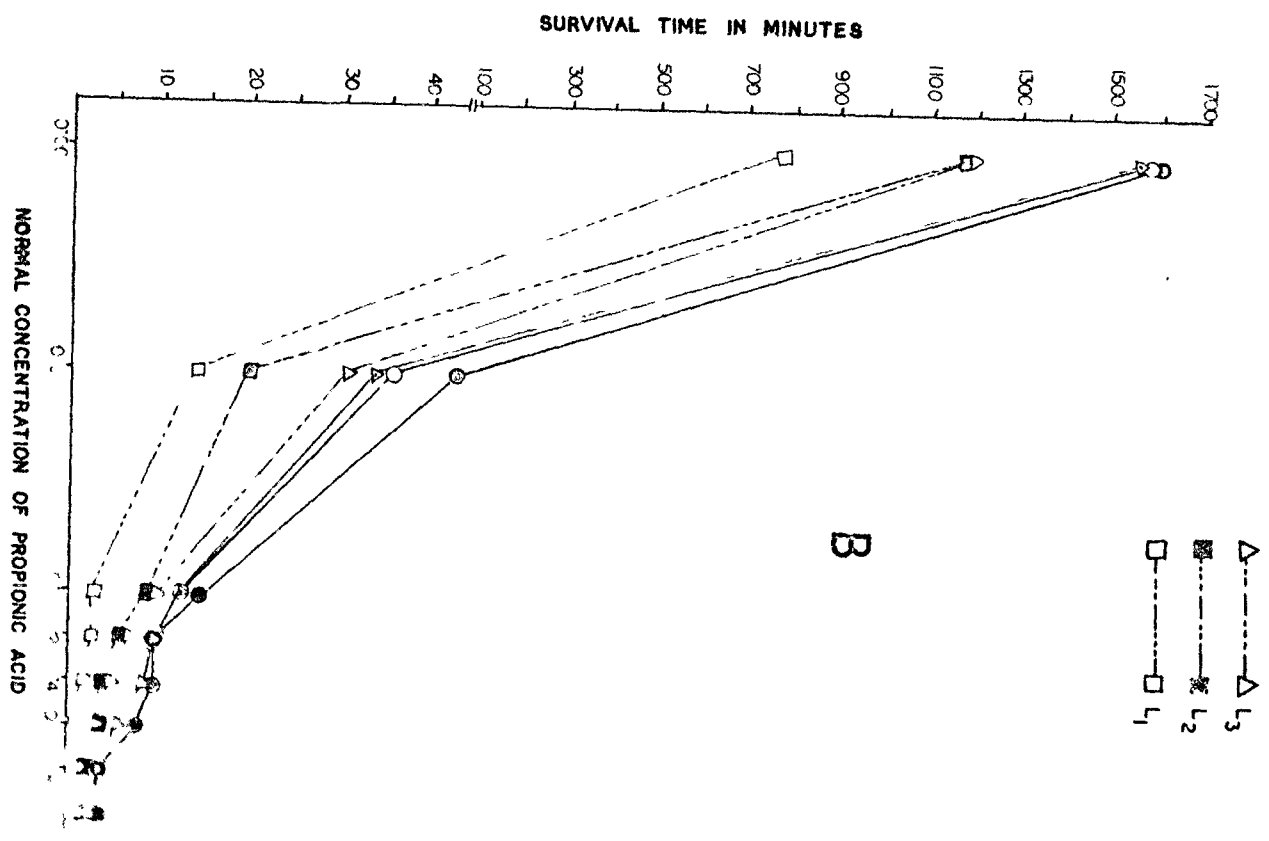
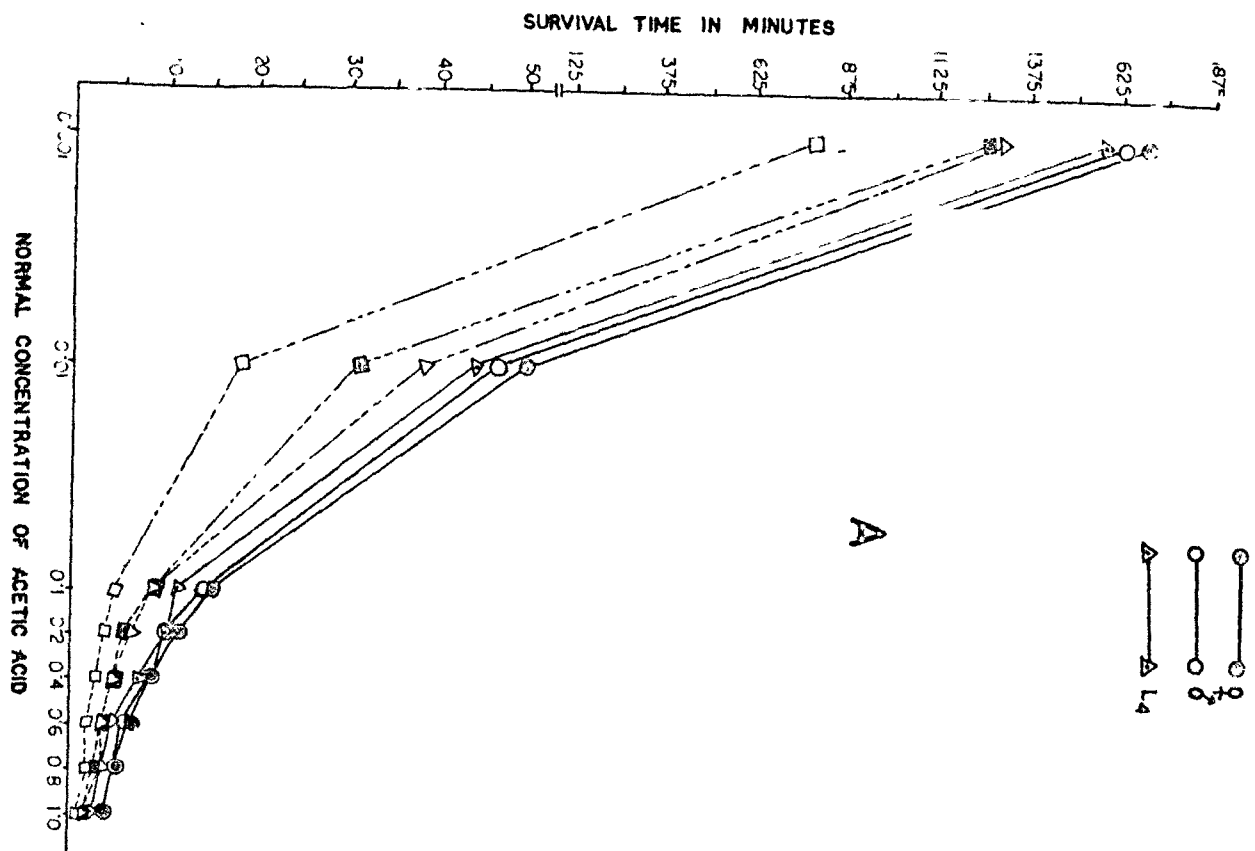


Fig. XXXVII

Survival time of adults and juveniles at different concentrations of acids. A - Butyric acid; B - Formic acid.



